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RESEARCH ARTICLE

Effect of some plant extracts against the greater wax moth *Galleria mellonella* L.

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Abstract: The present study was carried out to evaluate the biochemical effects of four plants extracts (*D. stramonium* L., *Hyoscyamus muticus* L., *Nerium oleander* L. and *Eucalyptus camalulensis* Dehn.) on the late instar larvae of the greater wax moth *Galleria mellonella* L. The results showed that treatment of *D. stramonium* L. 4% gave the maximum total mortality percentage (73.33%), while the minimum total mortality percentage was (13.33%) which recorded by *N. oleander* L. 4%. Biological parameters were studied under laboratory conditions, the life cycle decreased by increasing plant extract concentrations. The life cycle could be arranged in ascending order as follow: 58.33, 56.67, 62.67 and 69.00 day for the larvae treated with *D. stramonium* L. 4%, Sakran 4%, *E. Camalulensis* Dehn., and *N. oleander* L. 4%, respectively.

Key words: *Galleria mellonella*, Plant extracts, Biological parameters, Mortality percentage.

Introduction

The honeybee is the most useful insect to man. Honeybee is attacked by many diseases and pests which cause weakness of colonies and honey production. Like the Greater Wax Moths *Gallaria mellonella* L. which is the most serious pest of honeybee combs (Hachiro and David, 2000).

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The greater wax moth (GWM) *Galleria mellonella* (Lepidoptera:Pyralidae) is the most serious pest of honeybee wax combs in storage places and can cause substantial losses to combs, hive material and bees in beehives all over the world.

The natural compound with high insecticidal activity against the wax moth and with low toxic effects to honeybees is essential. This will enable their use in integrated pest management programs to control wax moth in honeybee colonies as well as in storage areas without contaminating honey bee products with pesticide residues. This work aimed to

determine the efficacy of *D.stramonium L.*, *Hyoscyamus muticus* L, *Nerium oleander* L., and *Eucalyptus camalulensis* Dehn. extracts against late larval instar of the greeter wax moth, *G. mellonella*, as natural control agents and to study their effect on some biological aspects of the insect.

Material and Methods

Preparation of Tested plants extracts

Tested plants were exposed to an extraction process according to the method described by Ellis and Hayes (2009). Each sample (100 g) was extracted by using acetone solvent. Dry ground materials were placed in flasks (1/2 liter), filled with acetone solvent and left for 24 hr. at room temperature. The flasks were plugged and shaken in an electric shaker, and then the suspensions were filtered through Whitman no.1 filter paper. The solvents were evaporated from the filtrate by leaving it at the room temperature to obtain crude extracts. Ten grams of each crude was dissolved in 100 ml acetone according to Roman *et al.,* (2009) to obtain a 10 % stock solution (w /v), which was stored under refrigeration until needed. This stock solution was serially diluted by the solvent as required for the bioassay tests.

Table 1. Source of the tested plants

English name	Scientific name	Family	Used part
Jimsonweed	*Datura stramonium L.*	Solanaceae	Leaves
Egyptian henbane	*Hyoscyamus muticus* L	*Solanaceae*	Leaves
Nerium	*Nerium oleander* L	*Apocynaceae*	Leaves
Camphor	*Eucalyptus camalulensis*	*Myrtaceae*	Leaves

Toxicity of tested plant extracts against G. mellonella larvae

Tested materials were applied as liquid formulations of acetone extraction, sprayed on the pieces of wax at Petri dishes (10 cm) under laboratory conditions, where 4.0% concentrations of each extract were prepared from the stock solutions (100%). Ten individuals of late instar larvae of *G. mellonella* were placed in each Petri dish, which contained 20g of small pieces of pure wax. One ml of the 4.0 % concentration was added to the dish above the larvae and closed with other dish to prevent escaping of larvae. Daily observations were done to count the number of dead individuals. All treatments were replicated three times. The LC_{50} and LC_{90} values were estimated from the toxicity lines of the tested materials according to Finney (1971).

Effect of tested plant extracts on some biological aspects of G. mellonella.

Effect of the four concentrations of each extract (0.5, 1.0, 2.0, and 4.0%) from the stock solutions (100%), on some

biological aspects of *G. mellonella* when applied as liquid fonnulations of acetone extraction sprayed on the pieces of wax at Petri dishes under laboratory conditions, was studied according to Pastagia and Patel, (2007). Ten newly hatched instar larvae of the greater wax moth were placed in each dish which contained 20g of small pieces of pure wax, and then 5ml of the least concentration of each tested material was added to the dish above the pieces of wax and closed with other dish to prevent escaping of larvae. Treatments were replicated three times. Different biological aspects; larval and pupal durations, adult longevity, ovipositional period, incubation period, life cycle, hatchability was recorded. Experiments were conducted under the laboratory conditions of 23±2 °C and 50±5% RH.

The collected data through different experiments were subjected to one way analysis of variance according to method of Gomez and Gomez (1984)**.**

Result and Discussion

The efficiency of plant extracts on greater wax moth.

Mortality percentage of the 5th instar larvae

Data of Table (2) indicate that the mortality % increased with increasing the time of exposure, reaching its maximum after 72 hours. The highest mortality (73.33 %) was obtained by *D. stramonium*, followed by *H. muticus* (56.67%). and *E. Camalulensis*. (30.00 %), while the lowest mortality (13.33 %) was recorded by *N. oleander*, and no mortality % was recorded by the untreated larvae. These results are in harmony with those obtained by Abbasipour *et al.* (2011) who showed that the mortality increased with increased concentrations of *D. stramonium* and exposure time. After 12 h, high increase in mortality was recorded. These results suggest that the extract of *D. stramonium* may be of high value in grain storage against *Callosobruchus maculatus*, especially in subsistence agriculture where the plants are locally available to farmers with little resources to meet the high cost of pesticides.

Malika *et al.* (2016) found that the synergistic extract of *N. oleander* caused a remarkable mortality rate of larvae of *Tuta absoluta* that exceeded 90%. *N. oleander* presented a low toxicity not exceeding 30% for the hydro-ethanolic extracts and 40% for the hydro-methanoic extracts.

LC_{50} and LC_{90} of plant extracts against the 5th instar larvae.

Table (2) and Fig. (1) indicate that LC_{50} and LC_{90} values of the used plant extracts for *D.stramonium* , *H. muticus, E. Camalulensis* and *N. oleander* were (1.61 & 11.90), (3.23 & 34.94), (9.22 & 70.60) and (18.52 & 114.06) g/L respectively.

Malika *et al.* (2016) found that after 24 hours of exposure to the test, the LD_{50} values were very high (above 50%) for hydro-ethanolic and hydro-methanoic extracts for the two plants used (*Nerium oleander* and *Ricinus communis*). However, the LD_{50} values were relatively low for the synergistic extract (19%). The results showed that second instar and first instar larvae are more sensitive to all used extracts. The third instar and fourth instar are the most resistant, except for the synergistic extract**.**

Table 2. Mortality % as well as LC_{50} and LC_{90} of the 5th instar larvae of *G. mellonella* treated with plant extracts of 4% concentration.

Materials	Mortality % after			Total	Mean	LC_{50}	LC_{90}	Slope ± SE
	24 h	48 h	72 h					
D. stramonium	13.33	23.33	36.67	73.33	24.44	1.61	11.90	1.47±0.38
H. muticus	3.33	20.00	33.34	56.67	18.89	3.23	34.94	1.24 ±0.40
N. oleander	0.00	6.67	6.66	13.33	4.44	18.52	114.06	1.62±0.90
E. Camalulensis	0.00	16.67	13.33	30.00	10.00	9.22	70.60	1.08 ±0.48
Control	0.00	0.00	0.00	0.00	0.00			

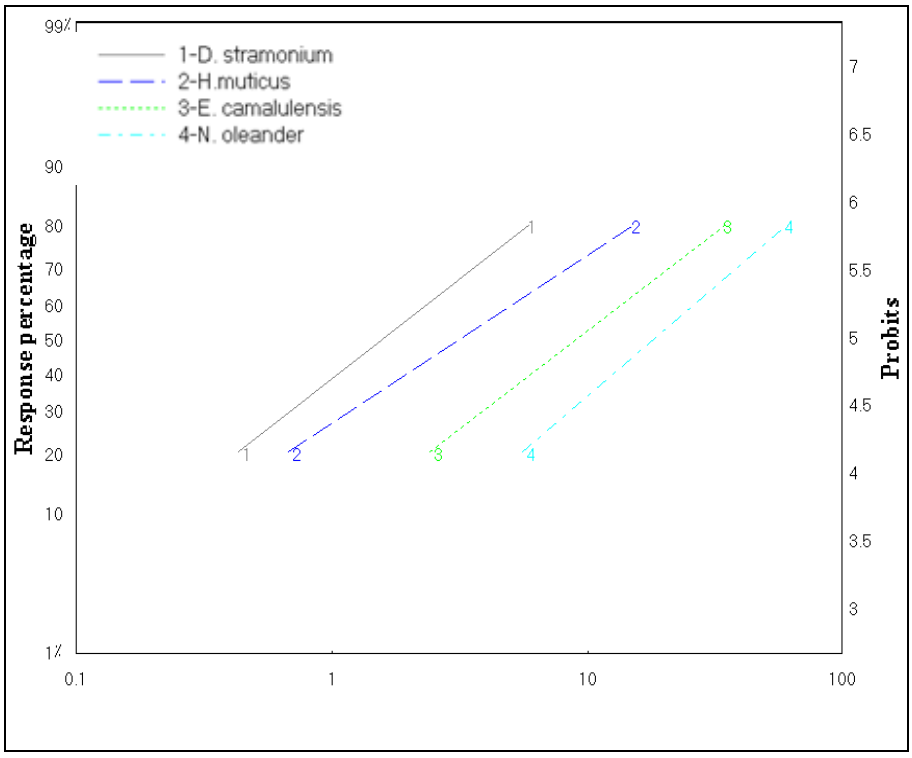

Figure 1. Bioassay of different plant extracts against the last larval instar of *G. mellonella.*

Effect of plant extracts on some biological aspects of G.mellonella under laboratory conditions.

Data presented in Table (3) and Fig. (2) describe the effect of different concentrations of *D. stramonium* on some biological aspects (larval and pupal periods, adult longevity, oviposition period, incubation period and consequently the life cycle) of *G.mellonella* under laboratory condition. The obtained results show that:

The duration of larval stage decreased by increasing *D. stramonium* concentrations (17.00 days at 4 % concentration, and 32.33 days at 0.5 % in comparison with 39.33 day at control). There were significant differences among all cases.

Pupal duration, adult longevity, and oviposition period also decreased by increasing *D. stramonium* concentrations. Statistical analysis proved that there were significant differences between adult longevity, and insignificant for the other two parameters.

On contrast, the incubation period increased by increasing *D. stramonium* concentrations, without significant differences.

The life cycle decreased by increasing *D. stramonium* concentrations, (58.33 days at 4 % concentration, 76.00 days at 0.5 % in comparison with 84.67 day at control). There were significant differences among all cases.

Effect of H. muticus L. against G.mellonella

Data presented in Table (4) and Fig. (3) describe the effect of different concentrations of *H. muticus* on some biological aspect of *G.mellonella*. The data show that:

The duration of larval and pupal stages, adult longevity, oviposition period and incubation period, decreased by increasing *H. muticus* concentrations. There were significant differences for larval and incubation period.

The life cycle decreased by increasing *H. muticus* concentrations (56.67 days at 4 % concentration, 76.00 days at 0.5 % in comparison with 84.33 day at control), with significant differences between concentrations and control.

Effect of N. oleander L against G. mellonella

Data presented in Table (5) and Fig. (4) describe the effect of different concentrations of *N. oleander* on some biological aspect of *G.mellonella* as follows:

The duration of larval and pupal stages, adult longevity, oviposition period and incubation period, decreased with increasing *N. oleander* concentrations. There were significant differences for all cases except for pupal and oviposition periods.

The life cycle decreased by increasing *N. oleander* concentrations, (69.00 days at 4 % concentration, 78.33 days at 0.5 % in comparison with 83.67 day at control), with significant differences between concentrations and control. **Effect of *E. Camalulensis Dehn. against G. mellonella***

Data presented in Table (6) and Fig. (5) describe the effect of different concentrations of ***E. Camalulensis*** on some biological aspects of *G.mellonella*.

Table 3. Effect of different concentrations of *D. stramonium* L. extract on some biological aspects of *G. mellonella* under laboratory conditions

***D.stramonium* Conc.**	**Duration period in days**					
	Larvae	**Pupa**	**Longevity of adults**	**Oviposition Period**	**Incubation period**	**Life cycle**
0.5%	32.33	13.67	13.67	5.67	11.67	**76.00**
2.00%	20.67	13.00	12.33	5.33	12.33	**63.33**
4.00%	17.00	12.00	11.33	5.00	13.00	**58.33**
Control	39.33	12.33	14.67	6.33	12.00	**84.67**
LSD 0.05	2.13	N.S	2.03	N.S	N.S	**3.44**

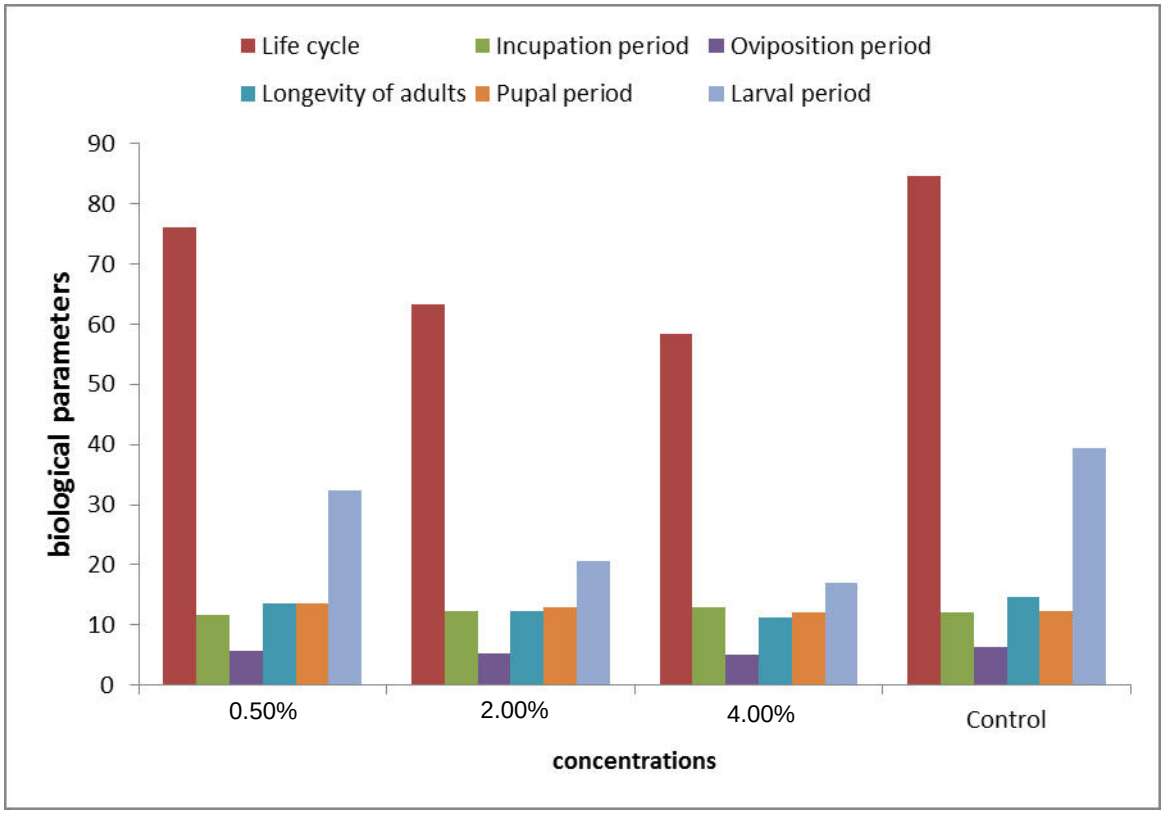

Figure 2. Effect of different concentrations of *D. stramonium* L. extract on some biological aspects of *G. mellonella* under laboratory conditions.

Table 4. Effect of different concentrations of *H. muticus* L. extract on some biological aspects of *G. mellonella* under laboratory conditions

***H. muticus* Conc.**	**Duration period in days**					
	Larvae	**Pupa**	**Longevity of adults**	**Oviposition period**	**Incubation period**	**Life Cycle**
0.5%	33.33	11.67	13.00	6.00	12.00	**76.00**
2.00%	23.67	11.33	12.67	5.67	11.00	**64.33**
4.00%	18.33	10.67	11.67	5.67	10.33	**56.67**
Control	39.00	12.33	14.33	6.33	12.33	**84.33**
LSD 0.05	2.21	N.S	N.S	N.S	0.67	**3.98**

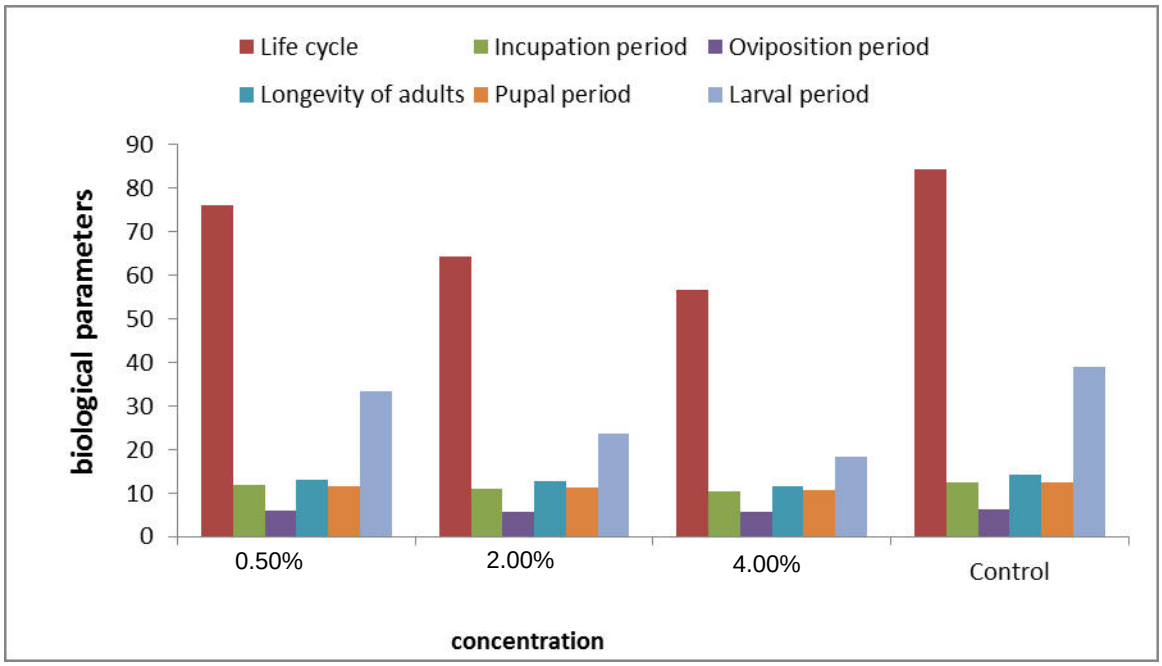

Figure 3. Effect of different concentrations of *H. muticus* L. extract on some biological aspects of *G. mellonella* under laboratory conditions

Table 5. Effect of different concentrations of *N. oleander* L. extract on some biological aspects of *G. mellonella* under laboratory conditions

***N. oleander* Conc.**	**Duration period in days**					
	Larvae	**Pupa**	**Longevity of adults**	**Oviposition Period**	**Incubation period**	**Life Cycle**
0.5%	34.33	12.33	12.67	6.67	12.33	**78.33**
2.00%	32.00	11.67	12.33	6.33	11.67	**74.00**
4.00%	29.33	11.33	11.67	6.00	10.67	**69.00**
Control	38.00	12.67	14.67	6.67	12.33	**83.67**
LSD 0.05	2.03	N.S	1.49	N.S	0.88	**3.72**

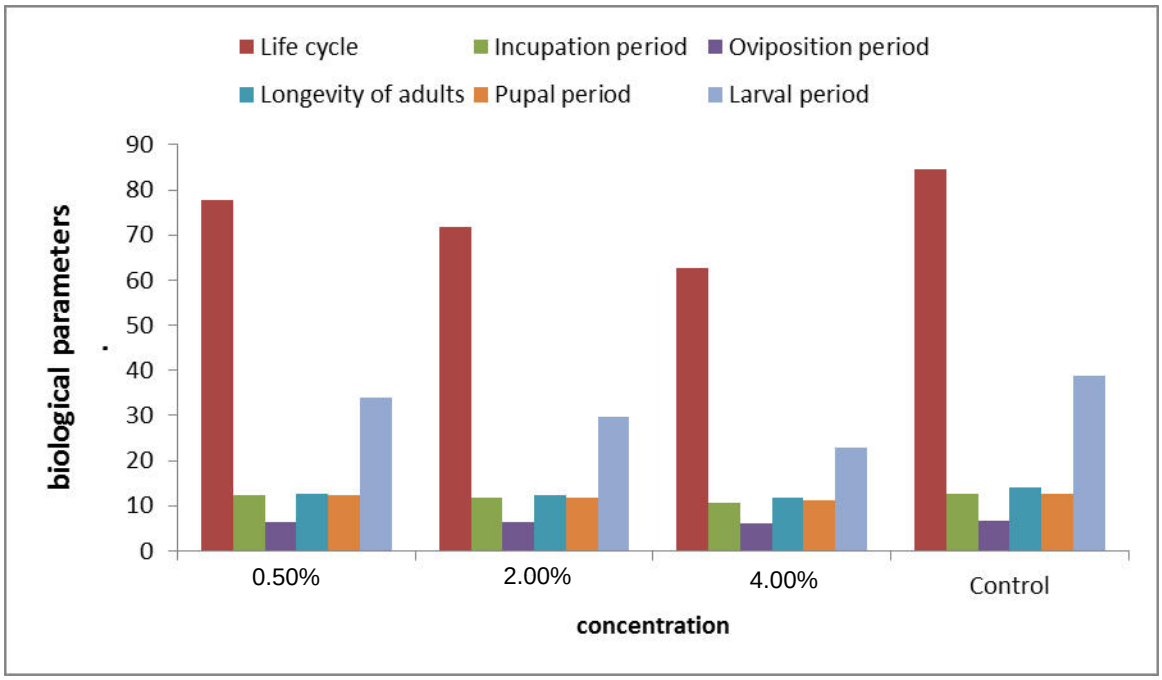

Figure 4. Effect of different concentrations of *N. oleander* L. extract on some biological aspects of *G. mellonella* under laboratory conditions.

Table 6. Effect of different concentrations of *E. Camalulensis* Dehn extracts on some biological aspects of *G. mellonella* under laboratory conditions.

E. *Camalulensis* Conc.	Duration period in days					
	Larvae	Pupa	Longevity of adults	Oviposition Period	Incubation period	Life Cycle
0.5%	34.00	12.33	12.67	6.33	12.33	**77.67**
2.00%	29.67	11.67	12.33	6.33	11.67	**71.67**
4.00%	23.00	11.33	11.67	6.00	10.67	**62.67**
Control	38.67	12.67	14.00	6.60	12.67	**84.67**
LSD 0.05	2.18	N.S	1.49	N.S	1.00	**4.16**

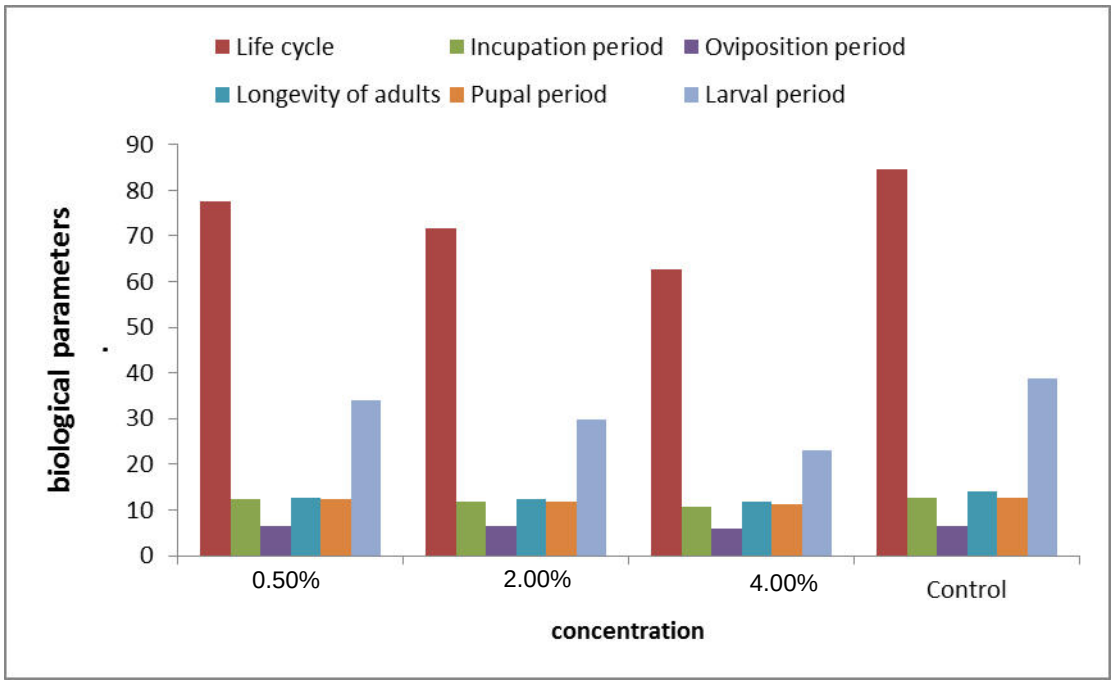

Figure 5. Effect of different concentrations of *E. Camalulensis* Dehn. extract on some biological aspects of *G. mellonella* under laboratory conditions

The duration of larval and pupal stages, adult longevity, oviposition period and incubation period, decreased by increasing *E. Camalulensis* concentrations. There were significant differences for cases except for pupal and oviposition periods.
The obtained results are in agreement with those found by Patel *et al.,* (1993). They found that *D. stramonium* L. plant gave higher mortality within 24 hr. against logicorn.

Kumral *et al.,* (2013) examined the ethanol extracts obtained from the leaf of the thorn apple (*Datura stramonium* L.) (Solanaceae) for lethal and repellent properties against adults of the European red mite *Panonychus ulmi* (Koch) (Acari: Tetranychidae) and its predator *Stethorus gilvifrons* (Muls.) (Col.: Coccinellidae) under laboratory conditions. The Petri leaf disc-spray tower method was used. The results showed that *Datura stramonium* leaf extracts were lethal to both the mite and its predator. Furthermore, an increase in the dose of leaf extract caused a significant increase in the death rates of both *Panonychus ulmi* and *Stethorus gilvifrons* adults.

Al-Ghannoum, and Karso, (2015) studied that Different concentrations (25, 30, 35 and 40%) of *Nerium oleander* L. leaf extracts were tested against the red grain beetle adults *Tribolium castaneum* (Herbst). Results showed that the percentage mortality ranged from 16.7% in the powder treatment increased to 70% in the alcohol extract at 40% concentration. Highest mortality rate was 49.2% in the alcohol extract, followed by 43.4% in the aqueous extract, while the least (30.9%) was recorded for the dry powder of N. oleander leaves. The results also showed an attraction and repellent effects of the Life cycle decreased by increasing *N. oleander* concentrations (62.67 days at 4 % concentration, 77.67 days at 0.5 % in comparison with 84.67 day at control), with significant differences between concentrations and control.
extracts on the pest adults and there was a positive correlation between high mortality rate of the tested products and repellent effects on the adults, while a negative correlation was recorded between mortality rate and attractiveness.

References

Abbasipour,H., Rastegar, F., Mahmoud, V.M. and Hosseinpour, M. (2011) 'Insecticidal activity of extract from Datura stramonium (F.) (Solanaceae) against Callosobruchus maculatus'. *IOBC/WPRS Bulletin; 2011. 69, pp.251-256.*

Al-Ghannoum, M.I. and Karso, B.A. (2015) 'Biological potency of *Nerium oleander* L. leaf extracts on mortality of the red flour beetle *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)'. *Egyptian Journal of Biological Pest Control; 2015.* 25(1), pp.135-138. 20 ref.

Ellis, A.M. and Hayes G.W. (2009) 'Assessing the efficacy of a product containing Bacillus thuringiensis applied to honey bee (Hymenoptera: Apidae) foundation as a control for *Galleria mellonella* (Lepidoptera: Pyralidae)', *J. Entomol. Sci*., 44(2), pp. 158-163.

Finney, D.J. (1971) 'Probit analysis', *(Cambridge Univ. Press, London, 3rd ed., 318 pp).*

Gomez, K.A. and Gomez, A.A. (1984) *'Statistical procedures of agric*', Res.John Wily and Sons, New York.

Hachiro A. and David A. (2000) *'Diagnosis of Honey Bee Diseases'*. U. S. Department of Agriculture ،Agriculture handbook No. AH –690, 61 pp.

Kumral, N.A., Cobanoglu, S. and Yalcn, C. (2013) 'Sub-lethal and lethal effects of *Datura stramonium* L. leaf extracts on the European red mite Panonychus ulmi (Koch) (Acari: Tetranychidae) and its predator, *Stethorus gilvifrons* (Muls.) (Col.: Coccinellidae)', *International Journal of Acarology*, 39(6), pp.494-501.

Malika, B., Badiaa, O. and Farida, S. (2016) 'Study of the larvicidal activity of hydro-alcoholic extracts *Nerium oleander* L. and *Ricinus communis* L. on *Tuta absoluta* M'. *VII International Scientific Agriculture* Symposium, "Agrosym, 6-9 October 2016.

Pastagia, J.J. and Patel, M.B. (2007) 'Biology of *Galleria mellonella L*. on brood comb of *Apis cerana* F. I. Plant Protec'. *Environ.*, 4 (2), pp. 85-88.

Roman, P.N.V. and Sera, B. (2009) 'Repellency and toxicity of three Impatlens species (Balsaminaceae) extracts on *Myzus percicae'*. *J. Biopesticides*, 2(1), pp. 48-51.