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INAPPETENCY AND LATENT EFFECTS OF VIRI-INFECTED LARVAE OF *Palpita unionalis* (HB.) [LEPIDOPTERA, PYRAUSTIDAE]

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ABSTRACT: The 2nd instar larvae of *Palpita unionalis* fed (under laboratory conditions of 26 ± 1 °C and 65 ± 3% R.H.) for 48 hrs. on newly vegetative olive leaves treated by dipping with different concentrations of aqueous extract of larvae of the same species infected with nuclear polyhedrosis viruses that were collected from laboratory mass-culture. The treated larvae high significantly consumed lower food than that concerning untreated ones during the periods of larval duration recording daily average of food consumption of 38.06, 42.75, 38.34, 35.11 and 45.30 mg/larva for the tested concentrations of 0.01, 0.005, 0.0025, 0.00125 and 0.00063 compared with 89.51 mg/larva for untreated larvae. Also, the daily averages of resulted faeces, utilized food and approximate digestibility high significantly varied according to the treatments. The biological aspects of larvae, pupae and emerged adults were influenced with treatments; where duration periods of both larvae and pupae of treated individuals prolonged; but mean weight of pupa and sex ratio as % emerged males reduced slightly, while % pupation, % emergence and longevity of both males and females reduced sharply comparing with that recorded for untreated ones.

Key words: ***Palpita unionalis, nuclear polyhedrosis viruses, food consumption, utilized food, approximate digestibility, duration, pupation, emergence, sex ratio, longevity.***

INTRODUCTION

The olive leaf moth, *Palpita unionalis* (Hb.) is considered as one of the phyllophagous-economic lepidopterous insects that mostly attack Oleaceae plants of genera *Olea, Jasminum, Ligustrum, Fraxinus* and *Phillyrea*

throughout the Mediterranean Basin and other countries. Its larvae attack the young shoots of olive trees, firstly, feeding off the parenchyma of the lower surface of leaves and finally devouring the leaves entirely including the major vein. In cases of particularly severe infestation, the larvae has been known to consume the pulp of green olives and moreover, attacks on young plants in the nursery can also be dangerous (Fouda, 1973; El-Kifl *et al.*, 1974; El-Sherif, 1975; Fodale *et al.*, 1990; Katsoyannos, 1992 and Pinto & Salerno, 1995).

Disturbances in the vitality of unhealthy insects take place in case of treatment with pesticides and/or infection with entomopathogenic viruses, bacteria, nematodes and fungi that cause certain pathological syndromes such as anorexia, dyspepsia and dormancy (El-Hakim & Hanna, 1982; Eid *et al.*, 1983; Abbas & El-Dakroury, 1988; Ahmed *et al.*, 1991; Zidan *et al.*, 1996; Tayeb, 1997; Lujuan *et al.*, 1999 and Hanna, 2004).

The present work aims to study the anorexial action on feeding behaviour and latent effects on certain biological aspects of the aqueous extract of viri-infected larvae of *P. unionalis* against the 2nd larval instar of the same species.

MATERIALS AND METHODS

Cadavers of larvae of the olive leaf moth infected with nuclear polyhedrosis viruses (NPVs) produced from laboratory mass-culture under 26 ± 1 °C. and 65 ± 3% R.H. were collected and kept in refrigerator. One gram of these cadavers was electrically blended in 100 ml of tap water for 3 minutes. Then, the suspension was sieved by a piece of muslin cloth to remove big particles. Afterthat, a series of gradual half-dilutions of 0.005, 0.0025, 0.00125 and 0.00063 was prepared from the obtained filtrate (as stock solution of 0.01).

Newly tender olive twigs were dipped in the abovementioned concentrations (in Petri dishes of 9 cm in diameter) for 5 seconds. Another twigs were dipped in tap water only for check individuals. For each concentration; 30 newly moulted larvae of the 2nd instar, in 3 replicates, were fed for 48 hrs. on

the treated air-drying leaves in Petri dishes (15 cm in diam.). Afterwards, the alive larvae for each replicate were transferred to another clean Petri dishes with untreated leaves for 48 hrs. intervals till pupation. The obtained pupae of each replicate were weighed and kept for emergence. The weights of both offered and residual leaves as well as resulted faeces were recorded every 2 days to estimate both ingested and utilized food in addition to the approximate digestibility according to Waldbauer (1968) as follows :

$$\text{Estimated food intake for larva} = \frac{\text{Wt. of offered food} - \text{food residues}}{\text{No. of larvae}}$$

$$\text{Daily consumption of food} = \frac{\text{Estimated food intake}}{\text{Feeding period (days)}}$$

Utilized food = Wt. of food ingested – Wt. of faeces

$$\text{Approximate digestibility (AD\%)} = \frac{\text{Utilized food}}{\text{Wt. of food ingested}} \times 100$$

Also, the longevity of larvae, pupae and emerged adults as well as % pupation, weight of pupa, % emergence and sex ratio were calculated.

The all obtained data were statistically analyzed with analysis of variance (F. test) according to Fisher (1944) and Snedecor & Cochran (1972).

RESULTS AND DISCUSSION

1.Food Intake and Faecal Matter

Data compiled in Table (1) indicate the mean weights of both consumed food and faeces of the 2nd larvae of *Palpita unionalis* fed on olive leaves treated with different concentrations of aqueous extract of viri-infected larvae of the same species for 2 days followed by untreated leaves (every 2 days) during the duration period. The mean weights of food consumed throughout the first period (feeding on treated leaves) high significantly differed according to the tested treatments recording the highest value (62.90 mg/larva) with the untreated control. The statistical analysis of variance revealed that the differences in the mean values of food consumption that ranged between 31.50 – 42.90 mg/larva were insignificant with the concentrations 0.01, 0.005, 0.0025, 0.00125 and 0.005, 0.00063. On the other hand, the variations in mean weights of resulted faeces/ larva during this period were insignificant having the range of 1.23 – 1.96 mg. During the 2nd period of duration, the food consumption for the tested

Table (1) : Food consumption and faeces of the 2nd larval instar of *P. unionalis* fed for 2 days on olive leaves treated with aqueous extract of viri-infected larvae of the same species and untreated leaves every 2 days during duration period.

Concent.	Mean weights of consumed food / faeces / larva (mg) at periods of 2 days interval											Daily average (mg)
	Treated	Untreated										
	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th	11th	
0.01	42.90 B	51.47	52.40 B	38.57 B	78.57 B	70.73 B	110.60 B	74.50	52.47 D	87.47 C	177.73 A	38.06 BC
	1.80	2.41 A	4.16	5.20	15.98	14.38	12.27	18.57	33.72	35.30 B	122.17 A	12.09 A
0.005	36.53 BC	43.87	47.20 B	37.70 B	64.80 B	58.37 B	94.37 B	80.60	97.20 A	159.60 B	220.20 A	42.75 B
	1.69	1.58 C	1.30	4.87	12.68	12.42	14.60	15.67	46.00	35.40 B	106.10 A	11.47
0.0025	38.60 B	46.30	49.67 B	39.70 B	67.30 B	60.57 B	92.70 B	136.87	81.40 B	123.20 B	107.17 B	38.34 BC
	1.24	1.36 C	4.69	5.87	9.14	14.89	15.50	16.23	37.40	33.50 B	45.30 B	8.42 B
0.00125	40.10 B	46.60	41.80 B	33.40 B	81.50 B	73.37 B	114.90 B	130.07	-	-	-	35.11 C
	1.23	1.37 C	2.24	4.39	22.24	20.02	16.27	11.70				4.96 C
0.00063	31.50 C	37.87	34.87 B	27.93 B	56.97 B	51.30 B	96.70 B	128.70	67.40 C	243.70 A	219.67 A	45.30 B
	1.73	1.77 BC	3.19	3.98	8.09	13.68	15.77	16.90	45.10	98.87 A	101.13 A	14.10 A
Control	62.90 A	67.77	100.20 A	111.30 A	318.70 A	270.97 A	321.27 A	-	-	-	-	89.51 A
	1.96	2.11 AB	8.24	12.86	20.10	11.17	11.80					4.87 C
F. test	**	N.S.	**	**	**	**	**	N.S.	**	**	**	**
	N.S.	**	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	**	**	**

treatments was statistically insignificant and ranged between 37.87 – 67.77 mg/larva. But, in case of the faecal matters, the reverse is true showing high significantly differences between treatments that recorded means weights of faeces of 2.41, 1.58, 1.36, 1.37, 1.77 and 2.11 mg/larva for the concentrations of 0.01, 0.005, 0.0025, 0.00125, 0.00063 and control, successively. With respect to both consumed food and resulted faeces for the 3rd, 4th, 5th, 6th and 7th periods of larval duration; statistical analysis of variance revealed high significantly (for the former) and insignificantly (for the latter) differences between treatments. In case of consumed food, the highly significant variations were shown only between the treated and untreated individuals, where the differences between the five tested concentrations were statistically insignificant. The untreated larvae consumed more food than that of the treated ones recording values of means weights of consumed food of 100.20, 111.30, 318.70, 270.97 and 321.27 mg/larva for the 3rd, 4th, 5th, 6th and 7th periods of larval duration, respectively. Whereas, the ranges of consumed food for the tested concentrations were 34.87 - 52.40 , 27.93 - 39.70 , 56.97 - 81.50 , 51.30 - 73.37 and 92.70 - 114.90 mg/larva for the same periods, consecutively. The insignificant differences of the resulted faeces of both treated and untreated larvae slightly varied and recorded ranges of mean weights of faeces of 1.30 – 8.24, 3.98 – 12.86, 8.09 – 20.10, 11.17 – 20.02 and 11.80 – 16.27 mg per larva for the 3rd, 4th, 5th, 6th and 7th periods of larval duration, successively. All individuals of control treatment pupated after about 14 days of treatment and both food consumption and resulted faeces of treated larvae insignificantly varied after 16 days of treatment showing ranges of 74.50 – 136.87 and 11.70 – 18.57 mg/larva for each, consecutively. But, after 18, 20 and 22 days the reverse took place, where the mean weights of both consumed food and resulted faeces of tested concentrations high significantly differed except with that of faeces that proved to be insignificant during the 9th period. The ranges of both consumed food and resulted faeces were 52.47 – 97.20, 87.47 – 243.70, 107.17 – 220.20 and 33.72 – 46.00, 33.50 – 98.87, 45.30 – 122.17 mg/larva after 18, 20 and 22 days of treatment, respectively.

Also, the daily averages of both consumed food and faeces high significantly varied according to treatments recording ranges of 35.11 – 89.51 and 4.87 – 14.10 mg/larva for each, successively. These results are confirmed with those obtained by Eid *et al.* (1983); Ahmed *et al.* (1991); Zidan *et al.* (1996); Tayeb (1997) and Hanna (2004) who reported that the food consumption and resulted faces of treated or unhealthy larvae of *S. littoralis, H. armigera* and *P. unionalis* were influenced due to treatments.

2. Utilization and Digestibility of Food

As shown in Table (2), the mean weights of utilized food significantly (at 1 and 5% levels of probability) vaired according to the tested concentrations, except with that of the 2nd, 8th and 11th periods of larval duration that proved to be insignificant. In general, the amounts of utilized food for untreated larvae were much more than that of treated ones. The mean weights of utilized food / larva for untreated individuals were 60.94, 65.66, 91.96, 98.44, 298.60, 259.80 and 309.47 mg, but that recorded for treated ones ranged between 29.77 – 41.10, 36.10 – 49.06, 31.68 – 48.24, 23.95 – 33.83, 48.88 – 62.58, 37.63 – 56.35 and 77.20 – 98.33 mg/larva during the 1st, 2nd, 3rd, 4th, 5th, 6th and 7th periods of duration of larvae, respectively. The duration of treated larvae prolonged for 8 days more than that of untreated ones recording ranges of utilized food of 55.93 – 120.63, 22.30 – 51.20, 52.17 – 144.83 and 55.57 – 118.53 mg/larvae during the 8th, 9th, 10th an 11th periods, consecutively. The differences in daily average of utilized food between the untreated and treated larvae were more highly significant, whereas that concerning of the treated ones were statistically insignificant. The values of daily average of utilized food per larva were 25.97, 31.28, 29.93, 30.15, 31.20 and 84.63 mg for the concentrations of 0.01, 0.005, 0.0025, 0.00125, 0.00063 and check successively. The results obtained by Eid *et al.* (1983) and Hanna (2004) stated that the amounts of utilized food of viri-infected larvae of both *S. littoralis* (the former) and *P. unionalis* (the latter) were significantly affected.

Respecting the approximate digestibility (Table, 2) there was no significance between treatments during the all periods of larval duration. Whereas, statistical analysis of variance showed high

Table (2) : Utilized food and approximate digestibility for the 2nd larval instar of *P. unionalis* fed for 2 days on olive leaves treated with aqueous extract of viri-infected larvae of the same species and untreated leaves every 2 days throughout the duration period.

Concent.	Mean weights of utilized food / larva and approximate digestibility at periods of 2 days interval											Daily average
	Treated	Untreated										
	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th	11th	
Utilized food / larva (mg)												
0.01	41.10 B	49.06	48.24 B	33.36 B	62.58 B	56.35 B	98.33 B	55.93	18.74 B	52.17 B	55.57	25.97 B
0.005	34.84 BC	42.29	45.90 B	32.83 B	52.12 B	45.95 B	79.77 B	64.93	51.20 A	124.20 A	114.10	31.28 B
0.0025	37.36 B	44.94	44.97 B	33.83 B	58.18 B	45.67 B	77.20 B	120.63	44.00 A	89.70 AB	61.87	29.93 B
0.00125	38.87 B	45.23	39.56 B	29.01 B	59.26 B	53.35 B	98.63 B	118.37	-	-	-	30.15 B
0.00063	29.77 C	36.10	31.68 B	23.95 B	48.88 B	37.63 B	80.93 B	111.80	22.30 B	144.83 A	118.53	31.20B
Control	60.94 A	65.66	91.96 A	98.44 A	298.60 A	259.80 A	309.47 A	-	-	-	-	84.63 A
F. test	**	N.S.	**	**	**	**	**	N.S.	**	*	N.S.	**
Approximate digestibility (AD %)												
0.01	95.97	95.29	92.73	86.93	79.79	79.79	88.65	61.60	35.71	53.11	30.23	68.01D
0.005	94.40	96.38	97.13	86.14	79.96	78.14	80.35	79.80	52.69	77.71	51.66	73.02 CD
0.0025	96.41	96.90	90.31	84.81	86.56	75.19	83.43	87.10	54.04	72.73	57.75	78.04 BC
0.00125	96.93	96.97	94.58	86.49	72.40	72.41	85.96	89.52	-	-	-	85.74 B
0.00063	94.51	95.30	87.39	80.40	84.47	71.66	83.07	86.33	35.62	58.69	51.96	68.90 D
Control	96.89	96.44	91.75	88.44	93.81	95.87	96.31	-	-	-	-	94.55 A
F. test	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	**

significantly differences in daily average of approximate digestibility between the examined treatments. The highest value of AD% (94.55%) was recorded with untreated individuals, whereas the lowest values of 68.01 and 68.90% were given in case of larvae treated with the concentrations of 0.01 and 0.00063 of aqueous extract of viri-infected larvae of *P. unionalis*. Many researchers such as Eid *et al.* (1983); Zidan *et al.* (1996); Lujuan *et al.* (1999) and Hanna (2004) found variations in approximate digestibility of unhealthy larvae of different insects.

3. Latent Effects

Data in Table (3) indicate certain biological aspects of treated individuals with the tested concentrations of aqueous extract of larvae of *P. unionalis* infected with nuclear polyhedrosis viruses compared with untreated ones. The studied biological aspects significantly and high significantly differed with treatments, except with differences of sex ratio that proved to be insignificant. The duration of both treated larvae and pupae prolonged recording ranges of 19.34 – 22.11 and 19.20 – 27.50 days for each compared with 14.31 and 13.77 days for untreated individuals, respectively. On the other hand, the pupation percentages, mean weights of pupa, % emergence, sex ratio as % emerged males and mean longevity of both males and females of treated individuals were more lower than that of untreated ones. The ranges of these aspects for treated individuals were 0.00 – 43.33%, 30.58 – 43.20 mg/pupa, 0.00 – 13.33%, 25.00 – 33.33%, 1.49 – 2.84 and 1.14 – 3.63 days comparing with 93.33%, 49.29 mg/pupa, 80.00%, 43.33%, 16.17 and 19.63 days for untreated individuals, successively. The fungi-infected larvae of *H. armigera* (Lujuan *et al.*, 1999) and viri-infected larvae of *P. unionalis* (Hanna, 2004) had the same latent effects.

Thus, the wastes and unhealthy individuals of insects produced from laboratory mass-rearing may be contained various pathogens such as viruses, bacteria, spores of fungi and nematodes that will be extracted and used as natural – safe biocide for injurous insects.

Table (3): Latent effects of aqueous extract of viri-infected larvae of *P. unionalis* on certain biological aspects of the same species.

Concent.	Mean of larval duration (day)	% Pupation	Mean of pupal duration (day)	Mean weight of pupa (mg)	% Emergence	Sex ratio as % emerged males	Mean longevity of adult (day)	
							♂	♀
0.01	22.11 A	26.67 D	19.20 BC	30.58 D	6.67 B	33.33	1.49 B	1.14 D
0.005	21.37 A	30.00 BC	27.50 A	37.47 C	10.00 B	25.00	2.30 B	2.53 C
0.0025	19.81 A	23.33 C	25.80 AB	40.90 B	0.00 B	-	-	-
0.00125	-	0.00 D	-	-	-	-	-	-
0.00063	19.34 A	43.33 B	25.00 AB	43.20 B	13.33 B	25.00	2.84 B	3.63 B
Control	14.31 B	93.33 A	13.77 C	49.29 A	80.00 A	43.33	16.17 A	19.63 A
F. test	**	**	*	**	**	N.S.	**	**

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فقد الشهية والتأثيرات المتأخرة ليرقات دودة أوراق الزيتون المصابة بالفيروس

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تم تغذية يرقات العمر الثانى لدودة أوراق الزيتون (تحت ظروف المعمل ٢٦ ± ١° م ، ٦٥ ± ٣% رطوبة نسبية) لمدة ٤٨ ساعة على أغصان زيتون طرفية حديثة غضة معاملة بالغمر فى تركيزات مختلفة من المستخلص المائى ليرقات نفس النوع المصابة بالفيروس والتى تم جمعها من التربية المعملية لهذه الحشرة. وقد استهلكت اليرقات المعاملة بدرجة عالية المعنوية كمية من الغذاء أقل منها بالنسبة لليرقات غير المعاملة خلال فترة حياة اليرقات حيث سجلت متوسط يومى للغذاء المستهلك ٣٨,٠٦ ، ٤٢,٧٥ ، ٣٨,٣٤ ، ٣٥,١١ ، ٤٥,٣٠ ملليجم / يرقة للتركيزات ٠,٠١ ، ٠,٠٠٥ ، ٠,٠٠٢٥ ، ٠,٠٠١٢٥ ، ٠,٠٠٠٦٣ مقارنة بـــ ٨٩,٥١ ملليجم / يرقة لليرقات غير المعاملة. كذلك اختلفت المتوسطات اليومية للبراز الناتج ، الغذاء المستفاد ومعدل الاستفادة بدرجة عالية المعنوية تبعاً للمعاملات. كما تأثرت الصفات البيولوجية لليرقات والعذارى والحشرات الكاملة بالمعاملة حيث استطالت فترة حياة كل من اليرقات والعذارى بينما انخفض قليلاً متوسط وزن العذراء والنسبة الجنسية مقدرة كنسبة منوية للذكور الخارجة، وانخفضت كثيراً نسبة التعذير، نسبة الخروج وفترة حياة كل من الذكور والإناث للأفراد المعاملة مقارنة بمثيلتها فى الأفراد غير المعاملة.