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EFFECT OF SOME ENVIRONMENTAL CONDITIONS ON DIAPUSE OF *LOBESIA BOTRANA* DEN AND SCHIFF (TORTRICIDAE : LEPIDOPTERA)

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Abstract

The combined effect of the three weather factors (temperature, photoperiod and relative humidity) on percentage of diapaused pupae of *Lobesia botrana* was estimated under field conditions. Percentage of diapaused pupae were highly influenced by daily means of both temperature and photoperiod, while relative humidity had no effect on active or diapaused insects. Long days (16 L : 8 D), especially when combined with high temperature, increased the rate of diapause development and decreased the time to subsequent adult emergence. Also, the exposure of diapaused pupae to low temperature (10ºC) for 15 days then reared at 25ºC and long photoperiod of 16 hrs. till emergence significantly shortened the pupal duration, but did not affect the pupal mortality. Diet and temperature may exert an effect on the percentage of pupae that enter diapause, however, these effects were not readily apparent except under short days less than 12 hours illumination.

INTRODUCTION

The ecology of insect diapause had been extensively studied, but most of the available data had been obtained for species evolved in temperature regions (Andrewartha, 1952; Lees, 1955; Beck, 1968). Such species were subjected to mark seasonal changes in photoperiod and temperature.

Experiments usually showed that photoperiod was the principal factor regulating diapause induction; while temperature influenced the range of inductive photoperiods. The relationship between climate and diapause of tropical insects had not been as thoroughly studied (Danilevskii, 1961).

Relatively minor seasonal changes in day length occurred in tropical and subtropical latitudes. These changes in day length, however, may serve to programme the development of some species (Norris, 1959; Zohdy and Abou El-Ela, 1975). Other climatic factors such as temperature and rainfall may play a more dominant role in regulating the onset and termination of diapause (Van Der Laan, 1959).

According to the previously mentioned discrepancies in literature, the present study was undertaken to obtain further data about environmental factors (temperature, food and photoperiod) which regulate pupal diapause induction and development of European grape berry moth, *Lobesia botrana* Den and Schiff.

MATERIALS AND METHODS

A. Diapause Induction: Four series of experiments were carried out at four different photoperiod conditions. These were; 0 hour light: 24 hours darkness (0L : 24 D), (8 L : 16 D), (10 L : 14 D) and (16 L : 8 D). In each series, numbers of newly deposited eggs were allowed to develop till the adult stage under the same light conditions. On hatching, groups of 25 larvae for each were kept in a petri dish and fed on fresh grape berries. These larvae were kept under continuous observation till pupation. Newly formed pupae were daily collected and soon after emergence the adults were counted. Pupae were considered in a state of diapause 30 days after pupation and percentages of diapaused and undiapaused pupae under the four tested photoperiods were calculated.

B. Diapause Termination: To test the effect of kinds of food (grape berries and lettuce leaves) in diapause termination, two series of newly deposited eggs were allowed to develop either on lettuce leaves or on grape berries and percentages of diapaused pupae were estimated.

The role of low temperature in diapause termination was also tested. Diapaused pupae were kept for 15 days at 10ºC, then kept at 25ºC and continuous darkness (0 L : 24 D) till emergence. The pupal duration of the diapaused pupae was calculated.

The effect of day length in diapause termination was also studied by comparing the percentage of diapaused pupae reared under long day (16 L : 8 D), intermediate (12 L : 12 D) and complete darkness (0 L : 24 D). Also, pupae were estimated under two degrees of temperature (18ºC and 25ºC).

The newly formed pupae, in the insectary allover the year and especially during October, November, December, January and February, were kept under the prevailing natural conditions of temperature and photoperiod till moth emergence and the mean diapaused pupal duration was then calculated.

The combined effect of the weather factors (temperature, relative humidity and photoperiod) during the diapause period (October – February) and the active period (March – September) was estimated by working out the partial regression (c-multiplier formula) adopted by Fisher (1950). The effect of daily means of temperature, R.H.% and photoperiod on moth emergence during both diapause and active periods was also estimated by using the correlation coefficient and partial regression values.

RESULTS AND DISCUSSIONS

A. The Induction of Diapause: In nature, diapause in *L. botrana* was restricted to pupae formed after the end of September that was determined by decreasing natural photoperiods shorter than 12 hours. On the other hand, it could be prevented by a gradual increase in photoperiod from 12 to 15 hours. The newly formed pupae in insectary during autumn/winter months (October – January) were kept under natural conditions of temperature, then photoperiod prevailing at that time of the year till moth emergence. Then, the mean diapaused pupal duration (30 days or more without emergence) was calculated, Table 1. The duration of pupae resulted from larvae pupated in insectary decreased from 132.6 days in October to 73.7 days in January under natural conditions prevailing at that time of the year. The same Table showed also that diapaused males emerged from pupae earlier than female moths.

1.The combined effect of three weather factors: The simultaneous effect of daily means of temperature, relative humidity and photoperiod on percentage of diapaused pupae of *L. botrana* was given in Tables 2 and 3.

It was found that the role played by the three weather factors was much pronounced during the first period (1^{st} October to 31^{st} March) where the three climatic factors were responsible for 88.87% of the total variance, Table 2. In the second period (from 1^{st} April to 30^{st} September), the three tested weather factors showed 53.14% explained variance Table 3.

a. Effect of daily mean temperature: Data in Tables 2 and 3 clearly indicate that the daily mean temperatures showed a highly significant value in the first period (October-March) which was characterized by low means of temperature, i.e. low temperature of that period was responsible for induction and maintenance of diapause. On the other hand, during the period from April-September, where the temperature was relatively high, this factor gave insignificant value, i.e. the high temperature of the period was responsible for normal active development.

Table 1. Duration of diapaused pupae and pupal mortality of *L. botrana* reared in insectary under natural conditions prevailing during winter months (October – January).

Months	Sex	Pupal duration in days			Average	%pupal mortality	Temperature (°C)			Mean photoperiod in hours / minutes	
		Min.	*Max*	Mean±SE			Min.	Max	Mean		
October	Male	36	179	124.1±7.8	132.6	13.25	24.70	27.43	26.07	11	41
	Female	50	179	141.2±6.7							
November	Male	31	177	116.7±4.8	123.5	8.43	20.40	22.36	21.38	10	49
	Female	31	162	130.3±2.8							
December	Male	43	128	107.7±3.7	111	7.25	17.33	18.78	18.06	10	18
	Female	65	131	114.3±3.4							
January	Male	31	98	69.6±4.3	73.7	11.11	10.50	17.8	14.15	10	21
	Female	55	98	77.9±2.6							

Table 2. Effect of daily means of temperature, relative humidity and photoperiod on percentage of diapaused pupae of *L. botrana* during the period of October-March.

Period	%of diapaused Pupae	Daily means of		
		Temp.(°C)	R.H%	Day length (min)
1-15 Oct., 1982	15.5	27.45	70.95	711
16- 31 Oct	23.7	26.50	71.25	690
1-15 NOv	33.9	25.63	70.80	659
16-31 Nov	42.3	23.28	72.28	639
1-15 Dec	60.2	22.27	74.13	622
16- 31 Dec	59.6	18.90	75.22	613
1- 15 Jan.,1983	53.0	18.92	72.85	616
16- 31 Jan.	53.0	16.93	78.15	625
1 -15 Feb.	52.0	16.77	74.90	646
16-28 Feb	47.7	17.98	76.98	670
1-15 Mar	45.9	17.83	69.31	691
16-31 Mar.	39.6	17.77	78.20	718
Simple correlation (r)		-0.76	+0.49	-0.76
Partial regression (b)		-2.04	-0.01	-0.22
Total explained variance (E.V.%)			**88.87%**	

Table 3. Effect of daily means of temperature, relative humidity and photoperiod on percentage of diapaused pupae of *L. botrana* during the period of April-September.

Period	% of Diapaused Pupae	Daily means of		
		Temp.(^{0}C)	R.H%	Day length (min)
1-15 Apr., 1983	18.7	20.02	70.53	750
16-30 Apr.	4.6	22.37	7023	771
1-15 May	0.0	22.48	65.93	801
16-31 May	0.0	25.33	69.14	819
1-15 Jun	0.0	27.62	75.93	837
16- 31 Jun	0.0	29.33	70.89	845
1-15 Jul	0.0	30.70	61.20	843
16- 31 Jul.	0.0	30.43	66.65	833
1- 15 Aug	0.0	30.68	71.20	816
16- 31 Aug.	0.0	29.98	73.27	794
1-15 Sep.	0.0	29.00	74.10	765
16-30 Sep	0.0	28.18	73.65	743
Simple correlation (r)		-0.70	+0.11	-0.50
Partial regression (b)		-0.84	-0.08	-0.04
Total explained variance (E.V.%)			53.14%	

b. Effect of daily mean relative humidity: The daily mean relative humidity yielded positive insignificant values. The partial regression was negative and insignificant in the first and second periods of the year. From these results, it could be concluded that the relative humidity had no effect on active or diapaused insects.

c. Effect of daily mean photoperiod (day length): The highest influence of the day length on the percentage of diapaused pupae of *L. botrana* was achieved in the first period (October – March), Tables, 2 & 3. Both correlation coefficient (r) and partial regression (b) values were highly significant during this period. On the other hand, this factor had no effect in the second period (April-September) i.e. the most important factor in induction of pupal diapause during the period (October-March) was the short day length prevailing during this time of the year. In this respect, Roehrich (1969) recorded that with *L. botrana* diapause happened in the pupal stage when half of the larval stage was spent under short day conditions, but photosensitivity was not limited to any particular part of the stage diapause occurred whenever the total number of days spent under short day conditions exceeded the total number spent under long day conditions.

B. The Termination of Diapause:

a. Effect of photoperiod: Table 4 showed that, at 25ºC, the average delay of emergence (pupal duration) under long photoperiod (16 L : 8 D) was 79.98 ± 1.7 days. It was 104.8 ± 6.4 days at intermediate photoperiod of 12 L : 12 D and 100.83 ± 2.4 at complete darkness (0 L : 24 D).

The time taken for 50% emergence at 25ºC was 81 days at long photoperiod and 106 days at complete darkness. Also, 101 days were needed for 90% emergence at long photoperiod and 123 days at complete darkness.

b. Effect of low temperature: Partial elimination of this diapause may be brought about at 10ºC for 15 days then transferred to 25ºC. The average delay of emergence at that condition was significantly short, i.e. 43.7 ± 11.3 days and 50% of the diapaused pupae could emerged after a delay of only 15 days after the exposure to low temperature (10ºC) for 15 days, Table, 5. On the other hand, the duration of exposure to low temperature was not significant and should be lengthened to several months at this temperature, for the time taken for 90% emergence at this condition was 123 days.

Table 4. Effect of photoperiods on diapause termination of *L. botrana* at 25°C.

Photoperiod	Pupal duration (in days)			Emergence		Mortality
	Min.	Max.	Mean ± SE	50%	90%	
16 L : 8 D	47	113	79.98 ± 1.7	81	101	30
12 L : 12 D	51	147	104.8 ± 6.4	96	119	22
O L : 24 D	57	170	100.8 ± 2.4	106	123	28.6
"F" value	6.2					
Significance evel : highly significant at 1%						

Table 5. Effect of temperature on diapause termination of *L. botrana*.

Temp.°C	Photoperiod	Pupal duration(in days)			Emergence		Mortality
		Min.	Max.	Mean ± SE	50%	90%	
25	16 L : 8 D	47	113	79.98 ± 1.7	81	101	30
18	16 L : 8 D	78	237	142.7 ± 4.1	149	184	17
10	O L : 24 D	1	151	43.7 ± 11.3	15	123	28
"F" value		6.3					
Significance level : highly significant at 1%							

Table 6. Effect of food on diapause termination of *L. botrana* under 25°C and 0L:24 D photoperiod.

Offered food	Pupal duration (in days)			Mortality %
	Min.	Max.	Mean + SE	
Grape berries	32	138	102.79 + 2.5	11.11
Lettuce leaves	32	135	81.52 + 3.8	20
"F" value	4.7			
Significance level : highly significant at 1%				

The percentage moralities at 25ºC was 30 and 28.6% under long photoperiod (16 L : 8 D) and complete darkness (0 L : 24 D). On the other hand, the exposure of diapaused pupae to low temperature for 15 days then reared at 25ºC till emergence did not significantly affect the pupal mortality, Table, 4. Relatively low temperature (18ºC) was found to be suitable for diapause because lower mortality (17%) was recorded and the delay of emergence was also considerably long (78-237 days) with a mean of 142.74 ± 4.1 days at this temperature.

c. Effect of food plants: Larvae were reared from the egg stage at 25ºC and green leaves of lettuce or grape berries were selected for testing the effect of food on diapause termination. The average pupal durations and percentage mortality associated with different types of food were recorded in Table 6.

From the Table it was obvious that the shortest pupal duration (81.52 ± 3.8 days) was associated with individuals reared on lettuce. They were subjected to slightly higher mortality during their pupal stage (20%).

The results of pupal duration of those reared from the egg stage on lettuce or grape berries were significantly analyzed and the difference in the average duration of pupal stage of both sexes reared on lettuce or grape berries was highly significant and it seems that food constituents offered to larvae play a role in diapause termination.

Perry *et al.* (1963) stated that diet and temperature may also exert an effect on the percentage of the population that entered diapause, however, these effects were not readily apparent except under short days. Under inductive photoperiods the greatest incident of diapause in the pink bollworm occurred in populations reared under the coolest temperature on diet having the greatest oil content.

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تأثير بعض الظروف البيئية علي طور السكون في دودة ثمار العنب

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قدرت نسبة عذاري دودة ثمار العنب (لوبيزيا بترانا) التي دخلت فترة سكون تحت الظروف الجوية الثلاثة (درجة حرارة - ضوء - رطوبة نسبية). أوضحت النتائج أن درجة الحرارة والضوء كان لهما تأثير كبير علي فترة سكون العذاري بينما لم تظهر النتائج أي تأثير للرطوبة. كما وجد أن تأثير الحرارة المرتفعة مع طول فترة الأضاءة (١٦ ساعة ضوء : ٨ ساعات ظلام) يزيد من نسبة فترة السكون للحشرات مع انخفاض في نسبة خروج الحشرات الكاملة. كما لوحظ انه عند تعريض العذاري التي في فترة سكون لدرجة حرارة ١٠°م لمدة ١٥ يوم ثم توضع علي درجة ٢٠°م وفترة ضوء ١٦ ساعة حتى خروج الحشرات وجد أن هذه الظروف قصرت فترة طول العمر للعذارى ولكنها لم تؤثر علي نسبة الموت في العذارى. وكذلك وجد أن لدرجة الحرارة ونوع الأكل تأثير كبير علي دخول الحشرات في فترة السكون الا أن هذا التأثير لا يظهر إلا تحت ظروف فترة الأضاءة القصيرة (أقل من ١٢ ساعة).