DIFFERENCES IN DEVELOPMENT OF STRAINS OF THE GYPSY MOTH

Porthetria dispar (L.)

David E. Leonard

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FOREWORD

The numbers of any organism in any one area fluctuate with time. The gypsy moth is an example of an animal whose numbers oscillate within a very wide range, from a few or no individuals per acre of woodland, to massive numbers whose quest for food results in complete defoliation.

There are many differing factors responsible for fluctuations in animal numbers. The ability to predict the trend is a prime asset in any sensible control program. Will the population numbers increase or decrease? Will areas now harboring low populations reach epidemic proportions in one, or several years? Should control measures be initiated, or will the population decline to insignificant levels without any external pressures?

With the gypsy moth, the number of egg masses per acre, and the size of these egg masses, provides some estimate of the potential damage (defoliation), but too often the predictions based on these data prove fallible.

Other factors might prove to be better criteria for estimating population trends. The recent development of an artificial food suitable for rearing the gypsy moth in the laboratory now provides an opportunity to examine biological differences in the gypsy moth. Future studies can determine their effects on the population dynamics of this insect.

The purpose of this study is to provide a reference point, or base, from which more detailed studies of the gypsy moth can proceed. This study shows that there are substantial differences in the segments of the populations sampled from various localities.

ACKNOWLEDGEMENTS

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Differences in Development of Strains of the Gypsy Moth

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INTRODUCTION

The recent development of a successful artificial food suitable for rearing gypsy moth larvae in the laboratory (Leonard and Doane 1966) has provided a better opportunity for detailed studies of this insect, including the re-study of some aspects of earlier investigations. Along with a reasonable degree of asepsis, this food assures a high percentage of adults from hatched eggs with a minimum of handling. The small amount of mortality, due primarily to diseases, provides little selective pressure on laboratory reared specimens.

Natural food was used for rearing this insect in earlier studies on development (Fernald 1896, Goldschmidt 1934, Ali 1933, and Maksimovic 1958). Because of difficulties associated with rearing on natural food, survival was usually low. Future laboratory rearings, both for biological studies and for control programs utilizing mass rearings, will utilize an artificial food. This investigation will establish a reference point for future use of artificial food.

The rate of development of *P. dispar* varies with temperature and humidity (Ali *ibid.*, Maksimovic *ibid.*), but it also varies with diet (Leonard and Doane *ibid.*, Leonard unpubl.). Rates of development of larvae reared on natural food and those reared on artificial food, therefore, may not be comparable.

Goldschmidt, in his extensive investigations, has shown many differences among populations of *P. dispar* from various geographic regions of the world. Aside from the brief work by Fernald and some study by Goldschmidt of a strain from Massachusetts, there has been no investigation of the development of North American gypsy moths, which ostensibly originated from a single introduction from France.

Goldschmidt shows conclusively that there is considerable variation in development time of different geographic races of gypsy moths. However, studies of variation within strains, essential elements of information for the study of population dynamics, are lacking. This investigation indicates that there is considerable polymorphism within the populations of this insect in North America.

MATERIAL AND METHODS

Egg masses were collected from the field and stored at 1-2° C for a suitable period of time to break diapause. Eggs from single egg masses were listed as strains, and labeled according to their geographic origin. LEB strains were from egg masses collected in Lebanon, Connecticut; BOZ, from Bozrah, Connecticut; and CAN, from Lacolle, Quebec, Canada. The series labeled CONN was taken from eggs collected in various locations in Connecticut, and being a mixture was not a strain.

Eggs were dehaired, and surface sterilized using a method described earlier (Leonard and Doane *ibid.*).

Rearings were conducted under differing conditions of temperature and light. The CONN series was reared at room temperature. Strains BOZ C and D, and a portion of LEB C were reared in a dark temperature cabinet maintained at 27 ± 0.5° C. A portion of LEB C was reared in a temperature cabinet with the same range of temperatures as above, but with 16 hours per day of illumination. The remainder of the insects were reared in a temperature controlled room at 23 ± 1.0° C and 16 hours per day of illumination. The temperature cabinets were kept in the controlled room. The relative humidity in this room ranged from 20 - 60%, depending on the season of year. No attempt was made to control the humidity in the petri dishes containing the larvae.

Newly hatched larvae were transferred with a camel's hair brush to a 100 x 100 x 20 mm plastic petri dish. Each larva was reared individually.

A block of food, approximately 25 x 25 x 10 mm, was placed on the inside of the top of the dish. This reduced contamination by frass which accumulated on the bottom. The food was placed in the corner of the dish towards the light to insure that the newly emerged larva would quickly find it. In the mixed strain CONN, however, the food was placed in the middle of the dish.

All handling of the larvae was performed in a disinfected transfer case to insure a reasonable degree of asepsis and reduce the incidence of larval diseases.

Individual data sheets were kept for each larva. Each insect was examined once daily, usually mid to late morning. The data were recorded for each change in life stage (molt, pupation, or emergence of adult).

Larvae were fed every third day until the last instars, when they consumed more food and were fed as necessary, sometimes daily.

Frass weights were used as a measure of feeding. In the CONN series, frass was collected daily and placed in a paper packet. The frass was dried in a dry heat oven for 1 hour at 150.5° C. The frass was then transferred to a weighing pan and weighed on a chain balance to the nearest 0.1 mg.

In strains LEB A, B, BOZ A, B, C, D, and E, frass was collected after the completion of a larval instar. The frass was placed in an aluminum foil packet and dried in a dry heat oven for 1 hour at 150.5° C. The packets were weighed, then emptied, and re-dried at the above temperature for 1 hour, then re-weighed. The difference in weight of the first and second weighings gave the weight of the frass. Handling of the foil and packets of frass was done with forceps.

Maksimovic (*ibid.*) has shown a daily loss in pupal weights. To insure uniformity for comparisons of weights of pupae, they were weighed on the day the pupa was formed, but only after tanning was completed. The pupae were weighed on a Metler H 4 scale to the nearest mg.

After weighing, pupae were kept in petri dishes lined with filter paper. The petri dishes were stored on edge. The filter paper provided a rough surface which facilitated eclosion of adults. When eclosion is hampered, malformed wings often result from hardening of the wings before extraction from the pupal case.

Prepupae were not differentiated as a distinct stage, and their development time was included in the last larval instar. The prepupal period lasts 3 to 4 days, during which no feeding takes place, and a coarse, sparse, silken cocoon is constructed.

RESULTS AND DISCUSSION

The first larvae studied were those in the CONN series. Several important points were learned from the individual rearings of these specimens, and these were confirmed in the more comprehensive studies that followed.

Rearing larvae individually greatly reduced the incidence of disease. In an earlier study, utilizing cultures containing relatively large numbers of larvae (Leonard and Doane, *ibid.*), the per cent mortality was 50.8%, in comparison to the mortality of 4.1% in this study. Individual rearing reduced the possibility of cross contamination by diseased larvae.

The average weight of pupae reared individually was higher than weights of pupae reared in mass cultures.

A large percentage of the CONN larvae underwent an extra larval molt. As seen in table 1, 26 of the 58 larvae reared from this mixture of eggs underwent an additional larval instar, i.e. 6 instars in males and 7 instars in females. The developmental times of extra molting larvae differed from what might be termed "normal" (5 instar male and 6 instar female) larvae. These four types of larvae will be called **5** males, **6** males, **6** females, and **7** females; the numeral denoting the number of larval instars.

These results, although unexpected, were found not to be unique. Fernald (1896) obtained from 17 female larvae, three **7** females, and from 21 male larvae, ten **6** males. This corresponds quite closely with the results obtained in the rearings of the CONN series.

Goldschmidt (1934) states that larvae may have 4 or 5 molts in both sexes, but makes no mention of 6 molts as one finds with females undergoing 7 instars. Goldschmidt's results show the number of instars as follows:

(1) both sexes with 5
(2) both sexes with 6
(3) all males 5, all females 6
(4) all males 5, females partly 5, partly 6
(5) males partly 5, partly 6, all females 6

These types, according to Goldschmidt, are found in definite geographic races. The type found in North America was with all males 5, all females 6 (type 3).

Table 1. Per cent of gypsy moth larvae undergoing an extra larval instar

Strain	Total Emerged	MALES N	MALES % 5 males	MALES % 6 males	FEMALES N	FEMALES % 6 females	FEMALES % 7 females
CONN.* (mixed)	53	34	55.9	44.1	12	63.2	36.8
LEB A	38	20	85.0	15.0	18	100	
LEB B	37	14	100		23	100	
LEB C	66	38	84.2	15.8	21	75.0	25.0
LEB D	27	8	87.5	12.5	19	100	
BOZ A	73	38	84.2	15.8	34	97.1	2.9
BOZ B	54	26	88.5	11.5	26	92.9	7.1
BOZ C**	68	31	83.9	16.1	35	94.6	5.4
BOZ D**	65	37	91.9	8.1	28	100	
BOZ E	58	26	84.6	15.4	25	78.1	21.9
CAN A	23	12	100		9	81.8	18.2
CAN B	23	17	82.4	17.6	6	100	
CAN C	20	12	91.7	8.3	8	100	
	552	279	87.5	12.5	252	92.3	7.7

* Reared at room temperatures.
** Reared at 27°C. All other rearings at 23°C.

Goldschmidt considered the number of molts dependent on a set of multiple alleles, T 1, T 2, and T 3. T 1 causes 4 molts in both sexes. T 2 causes 4 molts in males and 5 in females, and T 3 causes 5 molts in both sexes. The majority of races contain T 1 and T 2, or T 2 and T 3, and are frequently heterozygous.

In his study of Japanese strains, Goldschmidt (*ibid.*) found all of the five combinations listed above. However, the works of Nagasawa and Nakayama (1965) and Nagasawa (1957, 1965) have shown males with 5, 6, or 7 instars, and females with 6, 7, or 8 instars. No explanation for 7th or 8th instar female larvae is found in Goldschmidt's works, but it suggests that other alleles may be involved.

Since the CONN series contained eggs from a mixture of egg masses, larvae from single egg masses were reared individually to determine the incidence of extra instar larvae in single strains. Table 1 shows that in only one of the 12 strains examined, LEB B, were there no extra instar larvae of either sex. In LEB D, BOZ D, CAN B and C, females had no extra instar larvae, but in the CAN strain only small samples were available. Only in CAN A, as in LEB B, were there no extra instar male larvae.

The individual egg mass totals for extra instar larvae differed substantially from the mixed eggs in the CONN series (table 1). There were 55.9% of **6** males in CONN, whereas in the other strains 12.5% were **6** males. For females, the total of **7** females in CONN was 36.8% compared with 7.7% in the other strains. The total of extra instar larvae in CONN was 50.9%, and in the other strains, 10.1%.

There is a marked tendency for more males to undergo an extra instar than females, and if the 7th instar is governed by another allele of T there are undoubtedly modifying factors involved influencing its expression.

DEVELOPMENT OF LARVAE

A study of the development times for the various immature stages of the gypsy moth was undertaken to learn how much difference there was in development times of larvae undergoing an extra larval molt. In his comprehensive study, Maksimovic (*ibid.*) has shown that temperature has a great influence on the development time. In this study, 23 and 27° C were utilized to determine if temperature influences the incidence of extra molts. For **5** males the increase from 23° to 27° C causes an increase in development time of all immature stages ranging from 19.1 to 25.5%; for **6** males this increase is from 17.7 to 29.3%; for **6** females from 19.6 to 27.3%; and for **7** females from 3.2 to 60.3%.

Instar I

Table 2 shows that development time for **5** males varied by nearly 3 days at 23° C, but for **6** males the range was over 6 days. Extra instar male larvae took an average of over 1 day longer than **5** males to complete their development.

The range in variation of development time of females at 23° C was over 3 days, whereas the range in **7** females was 1½ days. The mean difference in females was nearly ¾ day, with the extra instar larvae taking longer to complete the first instar. Broad generalizations about **7** female larvae are not possible because of the small samples available for this study.

During this instar, males took slightly longer to complete their development than females.

Instar II

There was less variation in development time of this instar than in the previous instar (table 3). Extra instar larvae took longer to develop than "normal" larvae (excepting **7** females at 27°), but the difference in development times was less than in the first instar. Males completed the

Table 2. Average development time (days) of 1st instar larvae.
BOZ C and D reared at 27°C., remainder at 23°C.

Strain	5 males N	5 males Days	6 males N	6 males Days	6 females N	6 females Days	7 females N	7 females Days
LEB A	17	8.35	3	9.00	18	8.11	None	
LEB B	14	7.64	None		23	8.22	None	
LEB D	6	9.17	1	9.00	18	7.72	2	8.50
BOZ A	29	8.90	4	9.75	20	8.25	None	
BOZ B	21	8.48	1	10.00	19	8.21	None	
BOZ E	22	10.09	4	13.25	22	10.00	2	9.00
CAN A	13	8.77	None		9	9.11	3	10.00
CAN B	13	7.31	3	6.67	6	6.83	None	
CAN C	14	7.79	1	7.00	7	8.00	None	
	149	8.66	17	9.71	142	8.42	7	9.14
BOZ C	27	7.74	5	7.20	34	7.15	3	7.00
BOZ D	34	6.44	3	8.00	26	6.27	None	
	61	7.02	8	7.50	60	6.77	3	7.00

Table 3. Average development time (days) of 2nd instar larvae
(N same as in table 2)

Strain	5 males	6 males	6 females	7 females
LEB A	5.06	5.00	5.17	None
LEB B	6.36	None	5.17	None
LEB D	3.83	6.00	5.17	6.00
BOZ A	5.45	5.00	5.60	None
BOZ B	5.29	None	5.26	None
BOZ E	5.27	5.75	5.23	5.33
CAN A	4.85	None	4.44	4.50
CAN B	5.31	5.33	5.00	None
CAN C	4.29	7.00	4.29	None
	5.11	5.41	5.16	5.29
BOZ C	4.59	4.00	3.91	3.00
BOZ D	3.82	5.00	3.81	None
	4.16	4.37	3.87	3.00

2nd instar more rapidly than females, contrasting to the condition found in the first instar. Also, this instar is completed in 3 to 3½ days less time than the previous instar.

Instar III

With the exception of **7** females reared at 23° C, extra instar larvae had a shorter development time than "normal" larvae (table 4). Females complete their development sooner than males. This instar corresponds most closely to the 2nd instar in development time, but averaged about ½ day less time, excepting **5** males, which completed development about ½ day later than **6** males. **5** males took longer to develop than **6** females, and **6** males took longer than **7** females. **6** males, however, completed their development before **6** females.

Table 4. Average development time (days) of 3rd instar larvae
(N same as in table 2)

Strain	5 males	6 males	6 females	7 females
LEB A	6.00	5.67	5.44	None
LEB B	5.86	None	5.48	None
LEB D	5.00	3.00	5.06	7.00
BOZ A	5.66	4.75	5.70	None
BOZ B	5.76	6.00	5.91	6.67
BOZ E	6.50	6.00	5.91	6.67
CAN A	5.23	None	4.00	4.50
CAN B	5.23	5.33	5.00	None
CAN C	5.21	3.00	4.29	None
	5.71	5.18	5.36	6.14
BOZ C	4.52	5.00	4.53	2.50
BOZ D	4.68	2.67	3.81	None
	4.61	4.12	4.22	2.50

Table 5. Average development time (days) of 4th instar larvae
(N same as in table 2)

Strain	5 males	6 males	6 females	7 females
LEB A	7.41	5.33	6.00	7.00
LEB B	7.43	None	6.13	None
LEB D	7.00	5.00	5.39	7.00
BOZ A	7.72	5.25	6.20	None
BOZ B	7.81	5.00	5.26	None
BOZ E	7.41	6.00	6.32	6.67
CAN A	6.31	None	5.00	5.00
CAN B	6.54	5.67	5.33	None
CAN C	6.36	5.00	5.29	None
	7.24	5.47	5.87	6.29
BOZ C	5.56	4.00	4.91	2.50
BOZ D	5.35	5.33	4.42	None
	5.44	4.50	4.70	2.50

Instar IV

5 males took longer to complete their development than **6** males (table 5), the difference amounting to nearly 2 days at 23° and nearly 1 day at 27°. The development times of females are variable. **5** males took longer than **6** females, whereas **6** males took less time to develop than **6** females.

Instar V

Characteristically, the last larval instar of this species took about twice as long to complete its development as the previous instar. The last instar includes the time spent in the prepupal stage. Thus, the last instar of **5** males differs drastically from the 5th instar of **6** males, as shown in table 6. **6** males completed their development over a day before **6** fe-

Table 6. Average development time (days) of 5th instar larvae
(N same as in table 2)

Strain	5 males	6 males	6 females	7 females
LEB A	15.71	7.33	8.06	None
LEB B	15.86	None	8.00	None
LEB D	14.67	6.00	7.28	5.00
BOZ A	15.76	6.25	8.85	None
BOZ B	16.62	7.00	8.32	None
BOZ E	14.36	7.00	8.14	7.67
CAN A	13.77	None	6.44	5.50
CAN B	14.23	5.33	7.33	None
CAN C	14.64	7.00	7.57	None
	15.22	6.53	7.95	6.29
BOZ C	11.96	4.60	6.09	4.50
BOZ D	10.97	4.67	5.73	None
	11.41	4.62	5.93	4.50

males, but 7 females developed faster than both 6 males and 6 females. The development times for 6 males, 6 females, and 7 females are longer than in the previous instar.

Instar VI

At 23°, 7 females completed the 6th instar in about ½ the number of days that it took 6 females (table 7), but those few females completing 7 instars at 27° took longer than ½ the time of 6 females. The last instar of 6 males was shorter than the last instar of 6 females, and also, shorter than the last instar of 5 males. The development times for the last instar of 5 males and 6 females were nearly equal.

Table 7. Average development time (days) of 6th instar larvae and 7th instar larvae (N same as in table 2)

Strain	Instar VI			Instar VII
	6 males	6 females	7 females	7 females
LEB A	14.00	14.78	None	None
LEB B	None	15.17	None	None
LEB D	11.00	15.17	6.00	16.50
BOZ A	14.25	15.50	None	None
BOZ B	14.00	15.63	None	None
BOZ E	14.50	14.50	9.33	15.33
CAN A	12.33	14.44	6.50	12.50
CAN B	None	15.50	None	None
CAN C	14.00	15.57	None	None
	13.71	15.13	7.57	13.43
BOZ C	10.40	12.88	6.50	13.00
BOZ D	11.33	11.92	None	None
	10.75	11.00	6.50	13.00

Instar VII

The development time for 7 females (table 7) was slightly shorter than the last instar of 6 males at 23°, but considerably longer than 6 males at 27°, probably a reflection of the small sample size of 7 females. At 23°, 7 females completed the last instar more rapidly than the last instar of 6 females, but at 27° the opposite result was obtained.

Pupae

The most obvious feature of this stage was the shorter development time for females (table 8). At 23°, extra instar males and females completed their development before pupae of "normal" larvae, but the development of extra instar individuals at 27° was about equal to "normal" males and females.

Total development time of immature stages

The rate of development at both temperatures shows 5 males first (table 9) followed by 6 females about 3 days after, closely followed by 6 males, and last to complete their development were 7 females. The

Table 8. Average development time (days) of pupae (N same as in table 2)

Strain	5 males	6 males	6 females	7 females
LEB A	16.53	16.67	13.17	None
LEB B	16.57	None	13.30	None
LEB D	16.67	17.00	13.61	13.00
BOZ A	16.59	16.25	14.50	None
BOZ B	17.14	18.00	14.21	None
BOZ E	17.50	15.25	14.27	12.00
CAN A	16.62	None	13.00	13.50
CAN B	16.54	16.67	13.40	None
CAN C	16.57	16.00	13.29	None
	16.79	16.29	13.75	12.71
BOZ C	13.15	13.20	11.06	11.00
BOZ D	12.76	12.33	10.92	None
	12.93	12.87	11.00	11.00

number of days for 5 males, 6 males, 6 females, and 7 females were as follows: at 23° C, 58.77 days for 5 males, 62.59 days for 6 males, 61.65 days for 6 females, and 68.29 days for 7 females. At 27° C, in the same order as above, the number of days were 44.61, 48.75, 48.73, and 50.00.

Table 9 shows that there is much variation within strains in the total developmental times. For rearings at 23°, the range in the mean of days for 5 males is 6.27, for 6 males, 10.42, for 6 females, 7.97, and for 7 females, 10.00. The corresponding ranges for 27° are 3.50, 0.58, and, for 6 females, 1.85.

Variation in development times between strains can be seen most easily in figures 1 through 4. As could be expected, BOZ C and D, reared at higher temperatures, developed faster than the other strains at 23°. The three strains from Canada developed faster than the LEB or BOZ strains from Connecticut. The BOZ strains developed more slowly.

The faster development of the Canadian strains is quite evident. Goldschmidt (*ibid.*) noted that specimens from colder climates had a more rapid rate of development than those from warmer climates. If one can assume that the gypsy moths now present in North America originated from the single introduction from France, then this rather dramatic decrease in development time of specimens collected at the northern limits of the range in North America represents an adaptation to climate that has occurred in a comparatively short period of time, probably less than 50 generations.

Table 9. Average development time (days) from egg hatch to adult (N same as in table 2)

Strain	5 males	Difference from mean	6 males	Difference from mean	6 females	Difference from mean	7 females	Difference from mean
LEB A	59.06	+0.29	63.00	+0.41	60.78	—0.87	None	
LEB B	58.72	—0.05	None		61.48	—0.17	None	
LEB D	56.34	—2.43	57.00	—5.59	59.39	—2.26	69.00	+0.71
BOZ A	60.08	+1.31	61.50	—1.09	64.40	+2.95	None	
BOZ B	61.10	+2.33	65.00	+2.41	62.47	+0.82	None	
BOZ E	61.13	+2.36	67.75	+7.57	64.37	+2.72	72.00	+3.71
CAN A	55.55	—3.22	57.33	—5.26	56.43	—5.22	62.00	—6.29
CAN B	55.16	—3.61	None		58.39	—3.26	None	
CAN C	54.86	—3.91	59.00	—3.59	58.30	—3.35	None	
TOTALS	58.77		62.59		61.65		68.29	
BOZ C	47.52	+1.91	48.75	—0.35	48.73	+1.80	50.00	
BOZ D	44.02	—1.59	49.33	+0.58	46.88	—1.85	None	
	44.61		48.97		47.93		50.00	

Fig. 1. Development of 5 males of 11 strains of *P. dispar*. Even numbered larval instars are black, the pupal stage is stippled.

Fig. 2. Development of 6 males.

Fig. 3. Development of 6 females.

Fig. 4. Development of 7 females.

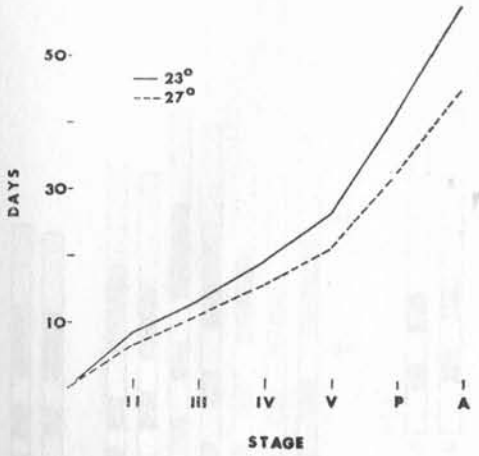

Figure 5. The effect of temperature on development of immature stages of **5** males.

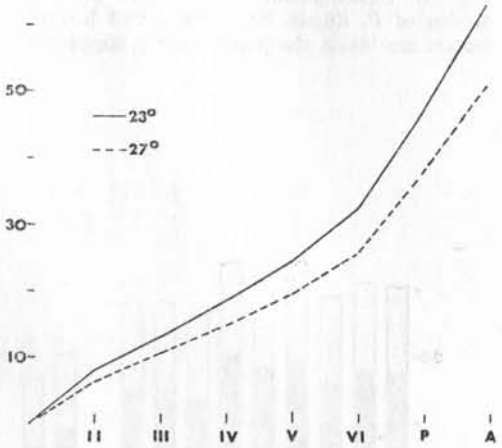

Fig. 6. The effect of temperature on the development of immature stages of **6** males.

EFFECT OF TEMPERATURE ON RATE OF DEVELOPMENT

The growth curves for the various types of larvae (figures 5 through 9) show a consistent decrease in development time for larvae reared at 27° C. This decrease in development time is, however, not due to a differential in development time in any one, or several, stages. Figures 9 through 12 show that the per cent of time spent in each stadium does not show much variation, regardless of the temperature at which the strain was reared. In **5** males (figure 9), most variation occurred in instars I, III, and V, with II, IV, and the pupal stage relatively stable.

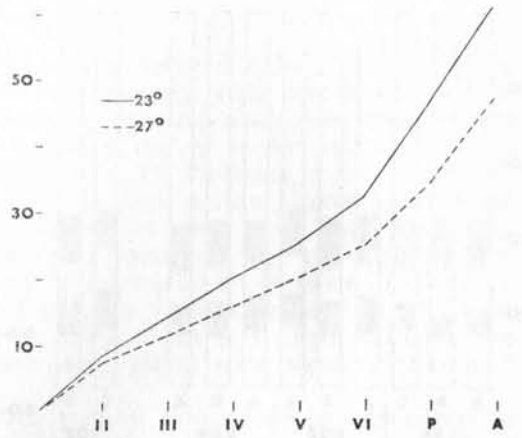

Fig. 7. The effect of temperature on the immature stages of **6** females.

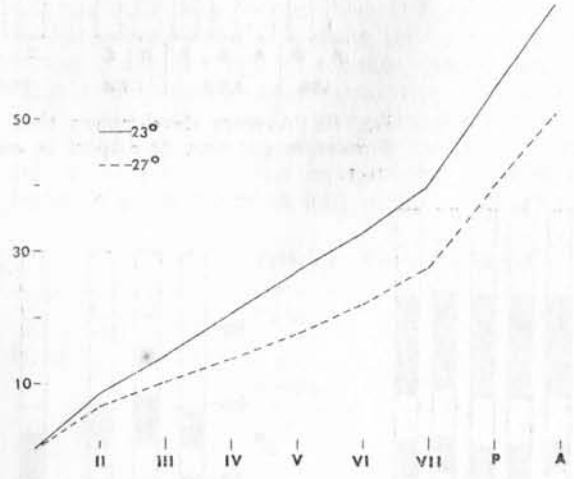

Fig. 8. The effect of temperature on the immature stages of **7** females.

In **6** males (figure 10) most variation in the per cent of time spent in each stadium occurred in I, II, V, and pupae. In **6** females (figure 11) instar I and the pupal stage were the most variable, and in **7** females (figure 12) there was variation in all but instar II.

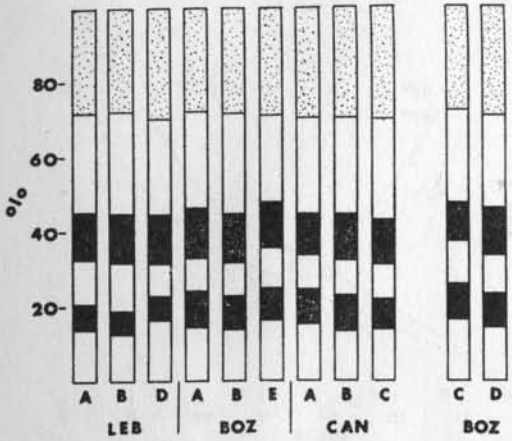

Fig. 9. Average development time of 5 males in per cent time spent in each stage.

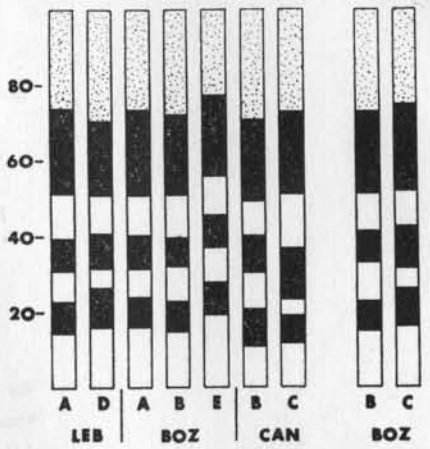

Fig. 10. Average development time of 6 males in per cent time spent in each stage.

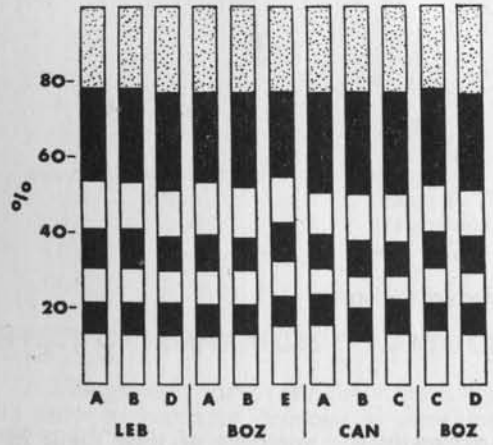

Fig. 11. Average development time of 6 females in per cent time spent in each stage.

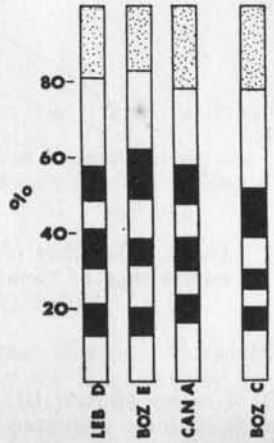

Fig. 12. Average development time of 7 females in per cent time spent in each stage.

DIFFERENTIAL PUPAL MORTALITY

The larval mortality for all strains averaged 4.1% (table 10) and it would be impossible to determine accurately what percentage of these might have undergone an additional larval molt, since most mortality was in instar I and V. Of the total mortality, the per cent in instar I was 46.4; II, 10.7; III, 7.1; IV, 10.7; V, 14.3; VI, 10.7. However, it was possible to determine the amount of mortality of extra molting larvae in the pupal stage. These data in table 10 indicate that there was a higher mortality in the pupae of larvae undergoing an extra molt. The totals in this table show that the pupal mortality of 5 males is 1.1%, compared to 8.0% in 6 males. The mortality in 6 females is 9.1%, compared to 19.3% in 7 females. In 6 females, however, 7.9% of the total pupal mortality can be attributed to BOZ A, where many pupae were abnormally formed and no adults emerged.

Since males with 6 instars have a slower rate of development, Goldschmidt considers T 3 inhibits the initial growth in males, making its growth more female-like, whereas T 1 makes female growth more male-like. This similarity in the growth pattern of 6 males and 6 females does exist (see figures 10 & 11). However, if the development of 6 males is to be considered more female-like, this condition only exists during the early instars, not during the last larval instar, which is decidedly more like the male than female. Also, it should be noted that sex is determined in this insect by the first instar, and can be determined by examination of larvae inside the egg (Levesque 1963). Sex reversal, if it occurs at a later time, should be accompanied by gynandromorphism, and no gynandromorphs were found in any of the rearings.

When the extra instar does occur, is it merely an additional molt following a normal last larval instar? Obviously not, for the last larval instar is about twice as long as the previous one, and extra instar indi-

Table 10. Larval and pupal mortality

Strain	N	Larval	Pupal			
		% larval mortality	5 males	6 males	6 females	7 females
CONN (mixed)	58	8.6	0	0	0	0
LEB A	43	11.6	0	0	0	None
LEB B	38	2.6	0	None	0	None
LEB C	68	2.9	0	0	0	0
LEB D	27	0	0	0	0	None
BOZ A	75	2.7	9.4	33.3	40.0	0
BOZ B	55	1.8	0	66.7	27.0	0
BOZ C	70	2.9	0	0	0	0
BOZ D	67	3.0	0	0	7.1	100
BOZ E	61	4.9	0	0	4.0	57.1
CAN A	26	11.5	0	None	0	None
CAN B	25	8.0	0	0	0	None
CAN C	20	0	0	0	0	None
	633	4.1	1.1	8.0	9.1	20.7

viduals do not undergo two prolonged instars at the end of their larval development. The larval stage most readily identified is the fourth instar when the head capsule is decidedly lighter than in the previous instars. Extra molting larvae have the characteristically lighter head capsule of "normal" larvae at 4th instar. Data from frass discussed later in the paper indicates the extra instar might occur at instar III or IV.

There is a method of determining whether last instar larvae will undergo an additional molt and whether or not a larva has undergone an extra molt. First, a larva in the fifth (male) or 6th (female) instar, if it is to molt again, will form a silken mat, and will stop feeding on about the 5th or 6th day, undergoing a period of quiescence prior to the molt. After the molt, the red tubercules characteristically found in late instar larvae undergo a slight color change, going from a dull brick red to a more brilliant red after the molt.

The lag in development of extra instar larvae is most apparent in the first instar. There is some suggestion that this lag is due to a behavior pattern that differs from that of other larvae, namely; that extra molting larvae spend a portion of their first instar wandering about the cage. During this wandering phase there appears to be no feeding or frass production. This aspect of the phenomenon of extra molting larvae is receiving further study.

The high percentage of extra instar larvae in the CONN series might be an effect of wandering. For these larvae, the food was placed in the middle of the dish. Many larvae did not find the food readily, and as their hunger increased, they became more active. Their movements were not random, but most larvae walked around the periphery of the dish, further delaying their finding of the food. To avoid this in subsequent rearings, the food was placed on the edge of the top of the petri dish nearest to the light. The larvae, attracted to the light, quickly found the food. After the first feeding, larval activity is greatly reduced.

Do extra instar larvae occur in the field, and if so, what is their effect on the population? These are questions that will receive attention in the future. It does not seem likely that extra molting larvae are unique to laboratory cultures, or a result of an artificial food. Fernald (1896) obtained similar results in rearing conducted in an insectary using leaves as food.

In some species of insects, males emerge prior to females, a condition known as protandry. The opposite condition, where females emerge before males, is known as protogyny. In field populations of gypsy moths in North America, protandry is most evident, but the selective advantage of this condition is not understood. The extra instar in males reduces protandry and, if there were populations consisting of large percentages of 6 males and 6 females, a shift to protogyny could occur. One might argue that the longer development time in males is necessary for the maturation of gametes, yet successful matings with males have been obtained within a few hours of the eclosion of males. There might be some advantage for later emerging males, since the longevity of males is shorter than females (Maksimovic 1958), and this might favor 6 instar males in the population. The occurrence of later emerging females would probably be disadvantageous, since there would be few if any males available, and this might explain the low frequency found in 7 instar

females both in this study and the results of Fernald. Much study is necessary before these points can be clarified.

EFFECTS OF ILLUMINATION AND TEMPERATURE

To determine whether the increase in development time in BOZ C and D was due to the effects of high temperatures or to the lack of illumination, a separate experiment was conducted, using individuals from one egg mass, (LEB C). The results are shown in figures 13 through 16, and in table 11. These data indicate the major influence on development time is temperature. Light may be exerting some influence, but additional data are needed to determine more clearly its total effect.

FRASS ACCUMULATION

The dry weight of frass was used as a measure of the amount of food consumed by larvae. Assuming that nutrient utilization was uniform, the larvae which consumed the most food should be the heaviest as well as producing the most frass. No attempt was made to determine the per cent utilization of nutrients.

Data from the CONN series indicated that extra instar larvae fed more and their pupal weights were higher. To substantiate this, frass was collected at the end of each instar from each larva from LEB A, B, BOZ A, B, C, D, and E. These data are included in tables 12 through 15. The frass weights for instars I and II are not included. Some indication of the dry weight of frass for these instars can be seen in table 18. The total dry weight of frass for the first two instars is about 5 mg or about 1/40th of the total for males, and about 1/160th of the total for females.

The frass production of instar III (table 12) shows 5 males with a slightly higher average production than 6 females, and 6 males with noticeably less frass than 5 males. The few 7 females produced more frass than 6 females. These data correspond well with the development time of instar III larvae (table 4).

In instar IV, 5 males again had the greatest frass accumulation (table 13). This corresponds to the longer length of time spent in this instar by 5 males. Quite surprising was the reduced amount of frass of extra instar larvae, both male and female, with 6 males producing an average of 37.5 to 40.0% as much frass as 5 males, and 7 females producing an average of 19.3 to 66.5% as much frass as 6 females. However, when one compares the figures obtained with "normal" 4th instars with extra molting larvae in the 5th instar (table 14), one can see that these figures are more closely alike. This suggests that the extra instar may occur during the middle of larval development, rather than at the beginning or end.

Most feeding occurred during the last larval instar. Table 14 shows that 5 males produce about four times as much frass during their last instar as during the previous instars. Also, 6 females began to feed more during this instar perhaps at a rate equal to or exceeding males, since their feeding lasted for a shorter period of time. The weight of frass produced by 6 males and 7 females in the 5th instar was nearly equal.

During the 6th instar, 6 males produced about the same weight of frass as last instar 5 males (table 15). Females produced a large quantity

Table 11. Average development time (days) of larvae from one egg mass reared under differing temperature and light regimes

# larval instars	Temp. °C	Illumination hrs.	N	I	II	III	IV	V	P	Total		
5	23	16	10	8.50	4.70	5.10	5.20	13.50	15.80	52.80		
Males												
	27	16	10	6.50	3.80	5.50	5.30	11.50	12.40	45.00		
	27	0	12	6.92	4.50	4.83	5.58	11.25	12.58	45.66		
				I	II	III	IV	V	VI	P	Total	
6	23	16	2	8.00	4.00	6.50	4.00	5.00	13.00	16.00	56.56	
Males	27	16	1	7.00	4.00	5.00	4.00	5.00	8.00	12.00	45.00	
	27	0	3	6.67	4.00	5.00	4.00	5.00	10.00	12.67	47.33	
				I	II	III	IV	V	VI	P	Total	
6	23	16	8	9.12	4.75	5.00	5.00	6.00	13.62	12.87	56.37	
Females	27	16	9	6.63	4.22	4.89	4.89	5.33	11.67	10.56	47.89	
	27	0	4	5.25	3.25	5.75	4.50	5.25	12.25	10.00	46.25	
				I	II	III	IV	V	VI	VII	P	Total
7	23	16	3	9.00	5.33	5.33	5.67	4.33	6.00	13.00	12.67	61.33
Females	27	16	1	8.00	4.00	3.00	3.00	6.00	4.00	13.00	13.00	54.00
	27	0	3	6.00	4.00	4.33	4.33	4.00	4.67	11.33	10.67	49.33

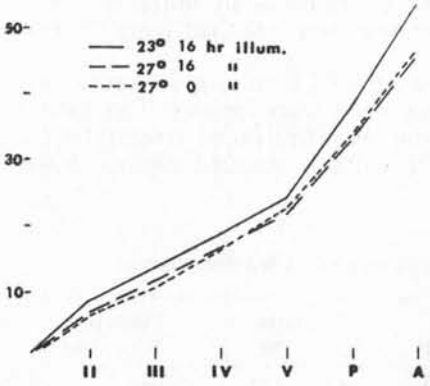

Fig. 13. The effect of light and illumination on development of the immature stages of **5** males.

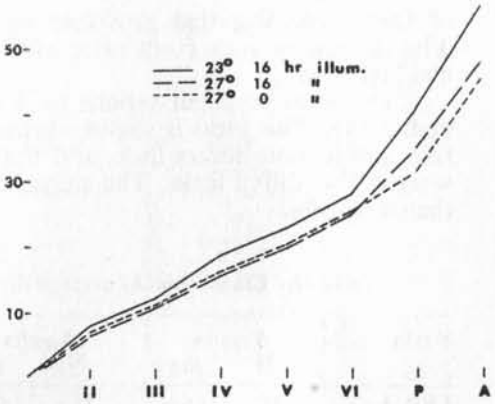

Fig. 14. The effect of temperature and illumination on development of the immature stages of **6** males.

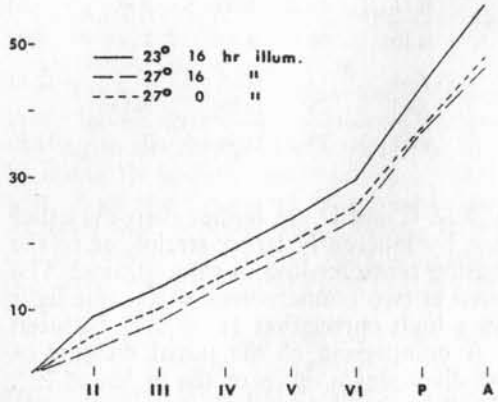

Fig. 15. The effect of temperature and illumination on development of the immature stages of **6** females.

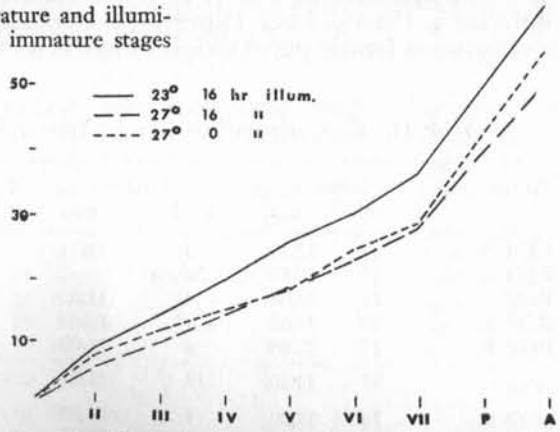

Fig. 16. The effect of temperature and illumination on development of the immature stages of **7** females.

of frass, exceeding that produced by last instar males by about 4-fold. The amount of frass from extra instar females was less than from "normal" females.

The ratio of pupal weight to dry weight of frass is quite uniform (table 16). This ratio is slightly lower in males than females. The total frass production differs little, and the ratios between pupal weight/frass weight also differ little. The pupae of **7** females weighed slightly less than **6** females.

Table 12. Comparison of average dry frass weights of 3rd instar larvae

Strain	**5** males		**6** males		**6** females		**7** females	
	N	mg	N	mg	N	mg	N	mg
LEB A	17	8.94	2	5.50	18	8.11	None	
LEB B	14	7.71	None		21	8.43	None	
BOZ A	24	9.37	4	6.00	18	8.72	None	
BOZ B	22	9.64	1	7.00	19	10.53	None	
BOZ E	22	7.95	3	6.33	22	8.00	3	9.00
	99	8.81	10	6.10	98	8.73	3	9.00
BOZ C	24	12.00	5	7.40	33	12.06	2	12.50
BOZ D	31	13.55	3	5.00	26	10.88	None	
	55	12.87	8	6.50	59	11.54	2	12.50

The dry weights of frass in BOZ C and D are higher than the other strains. Is this due to more feeding by inherently larger strains, or to the effects of higher temperatures causing more feeding in these strains? The results obtained with LEB C, reared at two temperatures, shed some light on this question. Table 16 shows a high correlation ($r = .89$) between frass weight and pupal weight. A comparison of the pupal weights of LEB C (table 17) shows the most divergent weights of the males, at 23°, 16 hours illumination, and at 27°, 0 hours illumination, are significantly different at the 5% level. However, the females show the opposite trend, and between female pupal weights there is no significant difference.

Table 13. Comparison of average dry frass weights of 4th instar larvae

Strain	**5** males		**6** males		**6** females		**7** females	
	N	mg	N	mg	N	mg	N	mg
LEB A	17	32.71	3	13.33	18	27.17	None	
LEB B	10	29.80	None		19	27.68	None	
BOZ A	25	34.96	4	13.00	19	29.26	None	
BOZ B	19	34.68	1	16.00	18	29.58	None	
BOZ E	22	28.86	4	13.00	21	22.96	3	18.33
	93	32.50	12	13.33	95	27.53	3	18.33
BOZ C	26	35.00	5	16.20	34	32.65	2	6.50
BOZ D	32	43.41	3	12.67	27	35.15	None	
	58	39.64	8	14.87	61	33.75	2	6.50

Table 14. Comparison of average dry frass weights of 5th instar larvae

Strain	**5** males		**6** males		**6** females		**7** females	
	N	mg	N	mg	N	mg	N	mg
LEB A	15	152.47	3	43.00	17	99.18	None	
LEB B	13	151.77	None		20	103.80	None	
BOZ A	27	172.04	4	41.75	20	123.10	None	
BOZ B	22	181.95	1	48.00	19	118.05	None	
BOZ E	21	147.95	4	34.25	22	102.95	3	40.00
	98	163.19	12	40.08	98	109.71	3	40.00
BOZ C	26	169.23	4	37.25	35	122.71	2	29.50
BOZ D	35	169.14	3	23.33	27	119.15	None	
	61	169.18	7	32.29	62	121.16	2	29.50

There is not a constant or uniform production of frass during any larval instar. Frass was collected daily from the CONN larvae. These data, in table 18, show frass was produced only about 80 to 90% of the time in days between molts. This is a reflection of the periods of inactivity before and after molting.

Table 18 also shows that there is a time lag in the development of extra instar larvae. In the first two instars it can be seen that corresponding to this lag in development is a reduction in days when frass was produced. In instar III and IV there is little difference in the percentage of days when frass was produced. Surprisingly, during the last larval instar, which includes the prepupal stage, during which there is no frass production, the percentage of days of frass production does not differ greatly from the other instars. The highest percentage of days when frass was produced is in instar I.

The average frass weights show the same trend as discussed earlier (tables 12 through 15), excepting that **7** females tend to show the same relationship to **6** females as **6** males show to **5** males. Because of the larger sample size of **7** females in CONN, this might be a closer estimate of the relationships existing between **6** and **7** instar females.

Table 15. Comparison of average dry frass weights of 6th and 7th instar larvae

Strain	Instar VI						Instar VII	
	6 males		**6** females		**7** females		**7** females	
	N	mg	N	mg	N	mg	N	mg
LEB A	3	160.33	17	610.65	None		None	
LEB B	None		22	620.23	None		None	
BOZ A	3	180.33	20	696.30	None		None	
BOZ B	1	192.00	19	743.68	None		None	
BOZ E	4	154.50	22	629.00	3	86.00	3	469.67
	11	166.54	100	661.40	3	86.00	3	469.67
BOZ C	8	184.80	59	744.37	2	127.00	2	694.00
BOZ D	3	112.00	24	695.00	None		None	
	11	157.50	83	724.29	2	127.00	2	694.00

Table 16. Total dry weight of frass (excluding instars I and II) and the ratio of pupal weight/frass weight, (r = .89) (all weights in mg)

Strain	5 males			6 males			6 females			7 females		
	Frass wt.	Pupal wt.	Frass/ Pupal	Frass wt.	Pupal wt.	Frass/ Pupal	Frass wt.	Pupal wt.	Frass Pupal	Frass/ wt.	Pupal wt.	Frass/ Pupal
LEB A	194	545	2.81	222	546	2.46	739	2215	3.00	None		
LEB B	189	550	2.91	None			770	2283	2.96	None		
BOZ A	216	594	2.75	241	664	2.76	857	2546	2.97	None		
BOZ B	226	508	2.25	263	673	2.56	902	2657	2.95	None		
BOZ E	185	552	2.98	208	589	2.83	763	2235	2.93	623	2005	3.22
BOZ C	216	607	2.81	246	680	2.76	912	2593	2.84	689	2669	3.07
BOZ D	226	661	2.92	153	416	2.72	860	2566	2.98	None		

Table 17. Pupal weight (mg) of specimens reared under differing light and temperature regimes

Strain	Temp. °C	Hrs. Light	N	5 males Pupal wt.	N	6 males Pupal wt.	N	6 females Pupal wt.	N	7 females Pupal wt.
LEB C	23	16	10	508*	2	461	8	2118**	3	2503
LEB C	27	16	10	529	1	503	9	2020	1	2223
LEB C	27	0	12	569*	3	548	4	2038**	3	2484

* Significant at 5% level using student t test.
** No significant difference using student t test.

Table 18. Dry weight (mg) of frass, duration of frass production, and pupal weights (mg) in a mixed strain of gypsy moth (CONN). The number of 5 males is 19, 6 males 15, 6 females 12, and 7 females 7

Type	Instar	Range days in stadia	Mean days in stadia	Mean days of frass prod.	% days of frass prod.	Range of frass wt.	Mean wt. of frass	Range of pupal wt.	Mean Pupal wt.
5	I	5-8	6.37	6.00	94.2	1.3-2.6	1.9		
6	I	6-10	7.47	6.20	83.0	1.5-2.6	2.0		
6	I	5-7	6.00	5.55	92.5	1.0-2.3	1.7		
7	I	5-10	7.00	6.00	85.7	1.4-2.5	1.9		
5	II	4-7	5.16	4.89	95.8	1.4-3.8	2.9		
6	II	3-14	6.07	5.47	90.1	1.8-3.9	2.8		
6	II	4-7	5.08	4.92	96.8	2.3-4.4	3.1		
7	II	5-9	6.71	5.29	78.8	1.5-3.0	2.4		
5	III	6-9	7.21	5.89	81.7	3.5-8.5	5.5		
6	III	6-8	6.93	5.73	82.7	1.9-4.9	3.3		
6	III	5-11	6.58	5.42	82.4	3.9-6.8	5.5		
7	III	6-8	6.85	5.57	81.3	2.6-6.6	3.5		
5	IV	7-9	8.42	7.28	86.5	14.5-31.4	21.5		
6	IV	4-7	5.67	4.80	84.7	6.9-12.0	9.1		
6	IV	5-9	7.00	5.92	84.6	8.4-23.7	14.3		
7	IV	5-7	6.00	5.14	85.7	7.7-12.0	10.4		
5	V	15-18	15.00	11.83	78.9	85.7-162.8	125.2	274-688	481
6	V	6-9	7.40	5.80	78.4	22.7-44.8	29.9		
6	V	8-9	8.33	6.83	82.0	46.2-91.8	68.5		
7	V	5-7	6.00	4.86	81.0	12.6-32.6	25.8		
6	VI	12-17	13.53	11.07	81.8	105.8-216.8	148.9	425-697	553
6	VI	15-18	15.83	12.83	81.8	379.9-596.7	435.8	1146-2171	1661
7	VI	6-8	6.85	5.71	83.3	70.0-103.0	84.7		
7	VII	13-17	15.29	13.00	85.00	390.8-621.8	516.1	1497-1983	1722

PUPAL WEIGHTS AND EMERGENCE

It was observed that the first pupae formed tended to be the heaviest. This observation is shown in table 19. The first individuals to complete their development, i.e. those adults emerging on the first two days of adult emergence, had pupal weights ranging from −1.5 to 21.8% heavier in males, and −2.5 to 24.0% heavier in females. Early emerging males in LEB A and B had lower average pupal weights, and those of BOZ B and CAN A were less than 5% heavier. In females, BOZ B and C weighed less, LEB D, BOZ E, and CAN B were less than 5% heavier. Only BOZ A, C, and D showed a consistent increase of over 10% in pupal weight of early emerging adults. LEB D, BOZ E and CAN B showed a greater increase in males than females, whereas BOZ D, and to some extent CAN A, showed the reverse trend. LEB A and B both showed slightly smaller pupal weights in early emerging males, but had higher than average pupal weights in females, with the reverse condition shown in BOZ B and CAN C. No strains showed both early emerging males and females with lower pupal weights.

If this differential in pupal weights occurs in the field, it could be of some consequence, especially in the case of females. Maksimovic (1958) has shown a direct relationship between pupal weights of females and the numbers of eggs produced by females emerging from these pupae. Heavier pupae yield females which lay more eggs. Faster developing larvae, at least in the laboratory, tend to be heavier, with a higher reproductive potential.

CONCLUSIONS

The purpose of this investigation was to establish a reference point or base for subsequent study of biological differences in the gypsy moth and their possible effects on population dynamics. Even in this preliminary study, substantial differences within and between strains are evident.

A small percentage of gypsy moth larvae of both sexes from Connecticut and Quebec, Canada, underwent an additional larval instar. Extra instar larvae were found in 11 of 12 strains, with a higher incidence of extra instar males than females. According to Goldschmidt, the variation in number of molts is controlled by three alleles of the T gene. The occurrence of seven larval instars in some females suggests that other alleles might be present.

Comparison of "normal" larvae showed that male and female development times were about equal in instars I and II, but males took longer than females to complete instars III, IV, V, and the pupal stage.

Extra instar larvae took longer to complete the first two instars than "normal" larvae, but all other immature stages proceeded at a rate faster than "normal" larvae.

The total development time from egg hatch to adult for **5** males was 58.77 days at 23°C and 44.61 days at 27°C, whereas **6** females took 61.65 days at 23°, and 48.73 days at 27°. This shows a marked tendency toward protandry. However, **6** males developed in 62.59 days at 23° and 48.75 days at 27°, closely approximating the times of **6** females. **7** females took the longest time to develop; 68.29 days at 23° and 50.00 days at 27°.

Table 19. Percentage difference in pupal weights (mg) of adults emerging during the first two days of emergence (5 instar males and 6 instar females only)

Strain	MALES					FEMALES				
	N 1st two days emerg.	% of total	Ave. wt. first emerg.	Ave. total pupal wt.	% difference	N 1st two days emerg.	% of total	Ave. wt. first emerg.	Ave. total pupal wt.	% difference
LEB A	5	29.4	537	545	−1.5	2	11.1	2752	2219	+24.0
LEB B	5	35.7	548	550	−0.4	8	34.8	2421	2283	+6.0
LEB D	2	33.3	608	499	+21.8	3	16.7	2199	2163	+1.7
BOZ A	3	10.3	661	594	+11.3	3	15.0	2829	2546	+11.1
BOZ B	2	9.5	631	598	+5.5	7	36.8	2590	2657	−2.5
BOZ C	8	29.6	702	611	+14.9	3	8.8	2606	2586	+8.8
BOZ D	2	5.9	728	662	+10.0	6	23.1	2743	2566	+23.1
BOZ E	3	13.6	607	552	+10.0	3	13.6	2257	2235	+1.0
CAN A	4	30.8	626	608	+3.0	3	33.3	2281	2152	+6.0
CAN B	3	23.1	615	509	+23.1	4	66.7	1918	1897	+1.0
CAN C	5	35.7	581	575	+1.0	5	71.4	2251	2271	−0.9

Larvae which underwent an additional molt tended to lag in their development during the first two instars, and this lag may have been due to the wandering habits of these larvae.

During what would normally be the last larval instar (V in males and VI in females), those larvae which underwent an additional molt had a differing behavioral pattern. Normally, in the last instar, larvae do not construct any silken webbing until the prepupal stage. Those which molt again construct a silken mat early in the stadia and cease feeding on the 5th or 6th day. The molt occurs in about a week after the instar begins, contrasting to a stadium of about two weeks if prepupae and pupae are to be formed. Also, the tubercules on the dorsum of **6** males and **7** females are a more brilliant red color.

Pupal mortality was highest in larvae which underwent an additional molt. An increase in temperature, from 23° to 27°C, reduced development time by nearly 25%. Lack of illumination had little, if any, influence on development time. The widest range in variation of development time occurred at 23°. The decrease in development times at 27° was not due to a differential rate in any one stage, for the per cent time spent in each stage did not differ from specimens reared at 23°.

The fastest developing strains were those collected in Canada. This suggests adaptation of these strains has occurred in a relatively small number of generations.

Using frass as an indicator of feeding, most feeding occurs during the last larval instars. The frass produced during instars I and II is about 1/40th of the total for males, and about 1/160th of the total for females. In instar III, **5** males produce slightly more frass than **6** females, but **6** males produced noticeably less frass than **5** males. In instar IV, extra instar larvae produced the least frass, with **5** males producing the greatest amount. In instar V, **5** males accounted for the most frass, but **6** females also fed heavily during this instar. Extra instar males and females produced about the same amount of frass. During instar VI, **6** females produced about 4-fold the amount of frass of last instar males. The last instar frass production of **6** males was equivalent to the frass produced by last instar **5** males, but **7** females produced less frass than **6** females during their respective last larval instars.

The ratio of dry weight of frass to pupal weight was quite uniform between strains, but slightly lower for males than females.

There was a tendency for the pupal weights of the earliest emerging adults to be heavier than those of later emerging adults.

LITERATURE CITED

Ali, Mithat. 1933. Experimentelle Untersuchungen über den Einflus ver Temperatur und Luftfeuchtigkeit auf die Entwicklung des Schwammspinners (*Porthetria dispar* L.) Zeit. fur angew. Entomol. 20: 354-381.

Fernald, C. H. 1896. The Gypsy Moth, *Porthetria dispar* (L.) Part II. Wright and Potter, Boston. p. 304-308.

Goldschmidt, R. 1934. *Lymantria*. Bibliographia Genetica 11: 1-185.

Leonard, D. E. and C. C. Doane. 1966. An artificial diet for the gypsy moth, *Porthetria dispar* (Lepidoptera:Lymantriidae). Ann. Entomol. Soc. Amer. 59: 462-464.

Levesque, G. 1963. A technique for sexing fully developed embryos and early-instar larvae of the gypsy moth. U. S. For. Serv. N. E. For. Exp. Sta. Res. Note NE 2: 1-3.

Maksimovic, M. 1958. Experimental research on the influence of temperature on the development and the population dynamics of the gypsy moth, (*Liparis dispar* L.). Posebno Izdanje Bioloskog Inst. NR Srbije 3: 115 pp. Trans. from Serbo-Croatian 1963. OTS 61-11203: 1-95.

Nagasawa, S. 1957. On the number of larval moults in the "Naheji" race of the gypsy moth, *Lymantria dispar* L. Problems on the breeding of insects for biological assay of insecticides 18. Botyu-Kagaku 22: 255-259.

——— 1965. Number of larval moults and the growth of the head capsule in successive instars in the "Shimizu" race of the gypsy moth, *Lymantria dispar* L. Problems on the breeding of insects for biological assay of insecticides 38. Kontyû 33: 466-474.

——— and I. Nakayama. 1965. On the number of larval moults in the Kyoto race of the gypsy moth *Lymantria dispar* L. Akitu 13: 8-13.