ACTIVITY PATTERNS AND ENERGETICS OF THE MOTH, *HYALOPHORA CECROPIA*

BY JAMES L. HANEGAN AND JAMES EDWARD HEATH

Department of Physiology and Biophysics, University of Illinois, Urbana, Illinois 61801

INTRODUCTION

The metabolic rate and fuel consumption of flying insects have been previously reported (Sotavalta & Lavlajainen, 1969; Zebe, 1954; Weis-Fogh, 1952; and others). The experiments of these workers were based on tethered insects flown to exhaustion on a known amount of fuel previously imbibed by the insect. The metabolic cost of flight over an insect's life span or the percentage of the total metabolism devoted to flight has not been reported.

The moth, *Hyalophora cecropia*, is ideally suited for an analysis of the energetics of flight. These heavy-bodied moths do not feed as adults (Rau, 1910) and both the substrate for flight muscle metabolism and the total energy reserve have been determined by Domroese & Gilbert (1964). They have a short adult life, averaging 1 week, and their activity can be monitored continuously.

For an analysis of the energetics of *H. cecropia* over its adult life the only variables that must be determined are the metabolic rate during various phases of activity and the total duration of flight. Thoracic temperature of *H. cecropia* has been correlated with activity (Hanegan & Heath, 1970). When the body temperature equals the air temperature the animal is inactive. Upon initiation of activity the thoracic temperature rises linearly to a precise temperature. During activity (flight and intermittent flight) the body temperature is regulated between 33·4 and 38·5 °C. Once the temperature difference is known an estimate of the metabolic cost of activity can be calculated (Heath & Adams, 1969).

MATERIALS AND METHODS

The experimental animal was the American silkmoth, *Hyalophora cecropia*. The moths were raised from eggs on wild cherry trees covered with large Nylon nets.

Temperature measurements. Calibrated thermocouples of copper-constantan were implanted along the dorsal mid-line of the mesothorax of both male and female moths. The thermocouple was glued in position with a small piece of cork and penetrated the dorso-longitudinal muscles to a depth of 1–2 mm. If an implant interfered with flight the animal was discarded. The leads of the thermocouple (4 mil diameter) were 1·5 m long and served as a tether. The animals were free to walk and fly within the limits of the tether length. The thoracic temperature of each animal was recorded on a Honeywell recording potentiometer (Electronik-16) at approximately 1 min intervals throughout the life of the animal. Nine to fifteen females were run at air temperatures

of 17, 25 and 30 °C. Nine and seven males were run at 18 and 25 °C, respectively. The light was controlled on a cycle of 16 h light and 8 h dark, approximating to the normal photoperiod at the time of emergence. The time of initiation and of the duration of activity (flight) was determined by monitoring thoracic temperature.

Oxygen consumption. Direct measurements of O_2 consumption in torpid and active moths were made with a Beckman paramagnetic oxygen analyser. The animals were placed in 125 ml flasks, and the flasks were immersed in a constant-temperature bath at 20, 25, 30 and 35 °C. The air temperature in the flasks and the thoracic temperature were monitored with thermocouples. The temperature data and the percentage decrease in oxygen concentration of a sample of effluent gas was recorded on a Honeywell recording potentiometer. The rate of oxygen consumption was determined by multiplying the percentage decrease in oxygen concentration by the rate of air flow (70 ml/min) through the system. The volume of air flow through the system was corrected to standard temperature and pressure. Values for the rate of oxygen consumption were used only if they remained stable for 3 min or more.

<center>RESULTS</center>

Analysis of cooling curves. Text-fig. 1 shows the thoracic temperature, monitored with respect to time, of a female moth at 25 °C air temperature. The animal's thoracic temperature equals ambient temperature when it is inactive; the moth warms up and maintains a high thoracic temperature when it is active. The duration of activity and the metabolic rate in calories per minute can be determined from this data; the duration is read directly from the graph, and the metabolic expenditure can be estimated from the rate of cooling when the moth ceases activity.
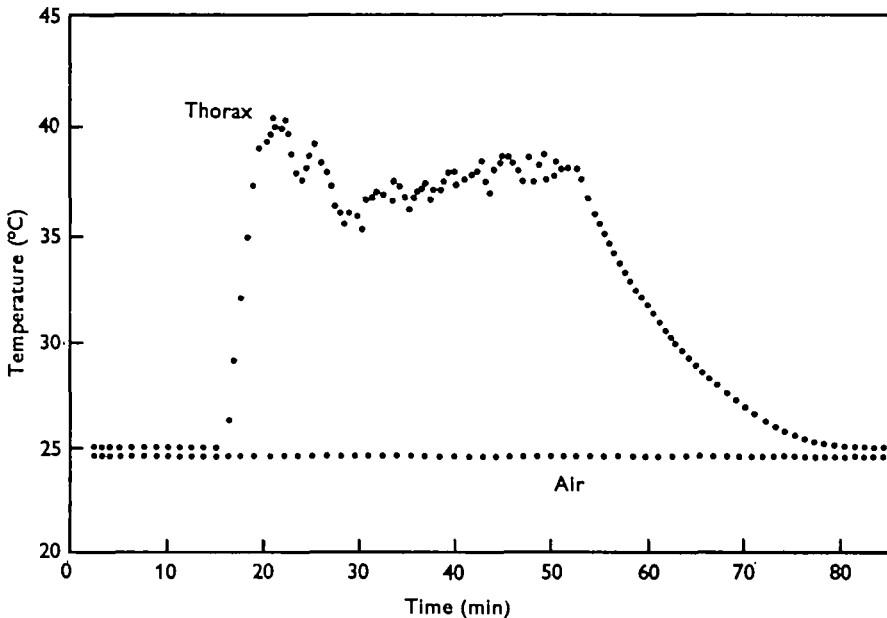
Text-fig. 1. Recording of thoracic temperature in a female moth at 25 °C air temperature.

The rate of cooling in these moths follows Newton's law of cooling which states that the rate of change in temperature (dT/dt) equals $K(T_B - T_A)$, where K is the combined constants for surface area of the cooling object and the thermal conductance, T_B is the body temperature and T_A is the air temperature. The rate of change in body temperature (dT_B/dt) can be read directly from the cooling curve for any value of $T_B - T_A + dT_B$. Two cooling curves were plotted in this manner for each animal to determine the best estimate of the rate of change in body temperature at the temperature gradient maintained during flight.

Table 1. *Effect of ambient temperature on the calculated metabolic rate during activity (flight) in* Hyalophora cecropia

Sex, air temp. (°C)	Cooling rate [K] °C/ min, °C difference	Rate of heat loss [dH/dt] (cal/min)	Temperature difference $[T_B-T_A]$ (°C)	Metabolic* rate [M] (cal/min)	Metabolic* rate corrected for convective cooling $[M_{AC}]$ (cal/min)	N
Male (18)	0·184	0·124	18	2·26 ± 0·14	2·66	14
Male (25)	0·160	0·085	12	0·98 ± 0·25	1·27	7
Female (17)	0·186	0·121	18	2·18 ± 0·54	2·72	11
Female (25)	0·179	0·095	12	1·30 ± 0·23	1·43	13
Female (30)	0·163	0·076	8	0·64 ± 0·12	0·76	15

* Mean ± 1 standard deviation.

When the body temperature is maintained at a constant value the rate of heat loss will equal the rate of heat production. The heat loss for any given value of dT_B/dt is given by; $dH/dt = WST_B/dt$, where dH/dt is the rate of heat loss, W is the weight of the cooling tissue (thoracic weight) in grams, and S is the specific heat in cal/g °C. The specific heat for insect tissue is 0·8 (Krogh & Zeuthen, 1941). Inserting the proper values into the above equation, heat loss, therefore heat production, at constant body temperature is given in cal/°C min. Multiplying this value by the magnitude of the established temperature difference (°C) yields the metabolic rate in cal/min of the individual moth during activity at the established temperature difference. This value is a minimum estimate, since it accounts only for the metabolic expenditure involved in heat production and does not include the metabolic output for flight or other physiological processes. Table 1 is a summary of these calculations for both males and females held at different air temperatures. The values indicated are means of the represented catagories; but, the metabolic rate (M and M_{AC}) were calculated on an individual basis and not on the reported means. K is the mean rate of cooling in °C/min °C difference and represents the slope of the cooling curve $(dT_B/dt)/(T_B - T_A)$. The heat loss (dH/dt) in cal/min °C was calculated from the general equation $dH/dt = WSK$. To determine the heat loss at constant body temperature, the heat loss per °C difference (dH/dt) was multiplied by the mean temperature difference $(T_B - T_A)$ established by the animal during activity. M represents the mean metabolic rate in cal/min to maintain a constant body temperature at the given temperature difference between the thorax and the air.

Effect of air flow on the cooling curves. Since the calculated metabolic expenditure in

actively flying animals is dependent on the rate of change in body temperature (dT_B/dt) in still air, a considerable error would result if air flowing over the animal, as in flight, significantly altered the rate of cooling. To study the effect of flowing air on the rate of cooling, four animals were anaesthetized and heated internally by passing current through a small coil of nichrome wire. The animals were placed in a wind tunnel in which the rate of air flow was calibrated with an Alnor thermo-anemometer. The cooling curves for individuals in different air flows were then plotted. A typical example of the cooling curves of one individual are shown in Text-fig. 2. There is an
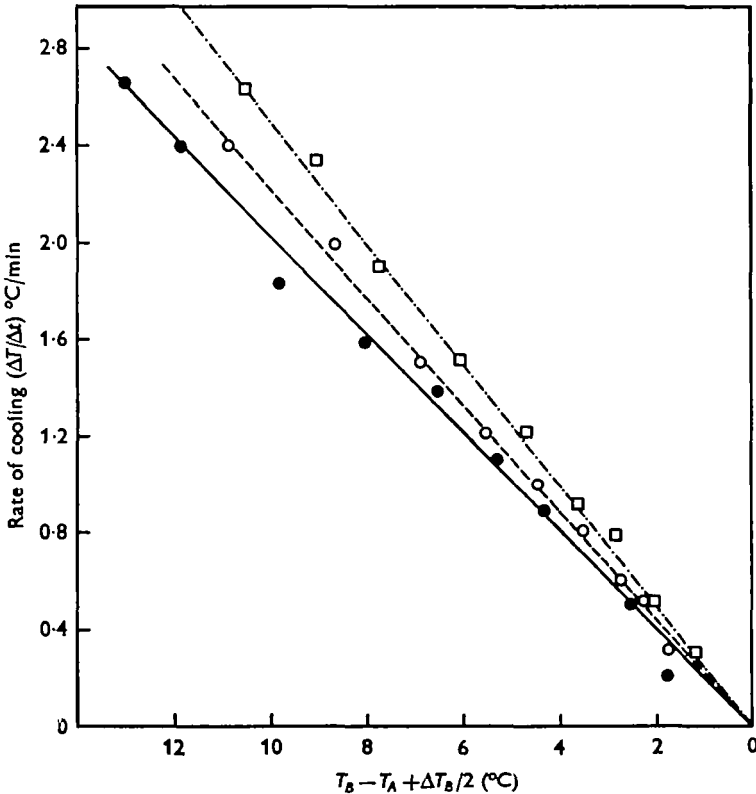

Text-fig. 2. Rate of cooling of a moth with respect to wind speed. ●—●, Still air; ○ --- ○, 0·54 m/sec air flow; □· · - · · -□, 0·85 m/sec air flow.

11 % increase in the rate of cooling in an air flow of 0·54 m/sec with respect to still air, and a 20 % increase in rate of cooling at an air flow of 0·85 m/sec. There was no further increase in rate of cooling at higher air speeds. The rate of cooling was measured in wind speeds up to 1·10 m/sec, which was the limit of the wind tunnel. The mean rate of increase of cooling in an air flow of 0·85 m/sec was 25 %. This increase in the rate of cooling due to the convective heat loss was added to the mean cooling rate (K), determined in still air. The calculations for heat loss using this new value of K were repeated and are listed in Table 1 as M_{AC}. M_{AC} represents the metabolic rate required of these moths to maintain a constant body temperature while in flight.

Metabolic rate from oxygen consumption. Oxygen consumption was measured

directly in both active and torpid animals. Assuming a caloric equivalent of oxygen of 4·8 cal/ml, the rate of oxygen consumption was expressed in cal/min. Text-fig. 3 shows the metabolic rate of torpid moths plotted against air temperature. In these animals the thoracic temperature equals the air temperature and both temperatures were monitored using thermocouples. The rate of increase indicates a normal Q_{10} response of 2·12.
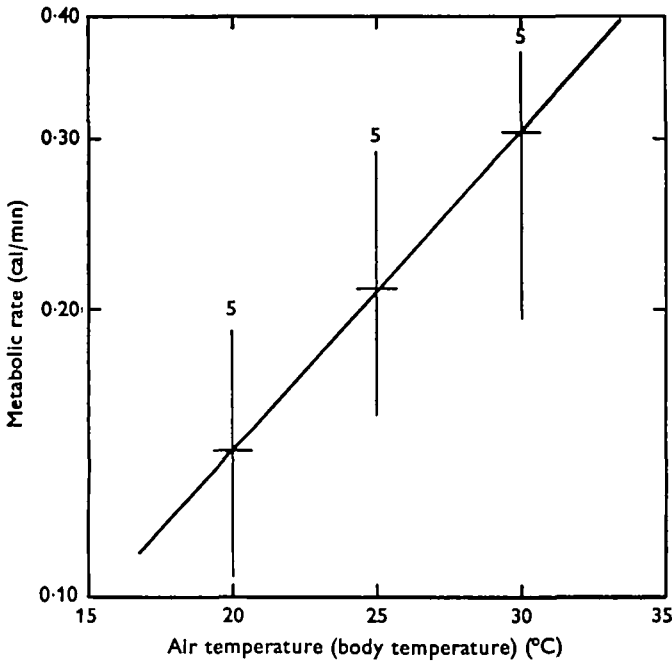

Text-fig. 3. Resting metabolism in torpid moths with respect to body temperature. Horizontal line = mean, Vertical line = range, Number = the sample number.

The metabolic rate of flying or active moths does not follow van't Hoff's rule. Rather, the metabolic rate increases with decreasing air temperature and is a function of the temperature difference established during activity between the thorax and air temperature. The thoracic temperature remains relatively constant during activity so that a decrease in air temperature causes the difference to increase. Text-fig. 4 shows this relationship of metabolic rate, air temperature, and temperature difference in active moths.

Table 2 is a summary of the metabolic rates of active and torpid males and females held at various air temperatures. M_{AD} is the measured metabolic rate (cal/min) of active animals and was determined directly from Text-fig. 4. M_T is the metabolic rate in cal/min of torpid animals and was determined directly from Text-fig. 3. The percentage difference between the calculated values of the metabolic rate (M_{AC}) and the directly measured rates (M_{AD}) during activity are indicated in Table 2. This percentage difference represents the metabolic expenditure of the animals for physiological processes other than heat production. At low air temperatures the greatest portion of the total metabolic output is for heat production. With increasing air temperatures the percentage of the total metabolic output for processes other than

heat production increases, up to 40% in females held at 30 °C. When the resting metabolism is added to the calculated metabolism for activity $(M_{AC}+M_T)$ the percentage difference found between this corrected value and the directly determined metabolic rate (M_{AD}) is zero in both the females and males held at 17 °C and 18 °C air temperature. The sum of the calculated metabolic rate and the resting metabolism does not include the calories converted to the mechanical output of flight or those calories lost through evaporation. It would therefore appear that flight would be impossible for the average moth at these air temperatures.
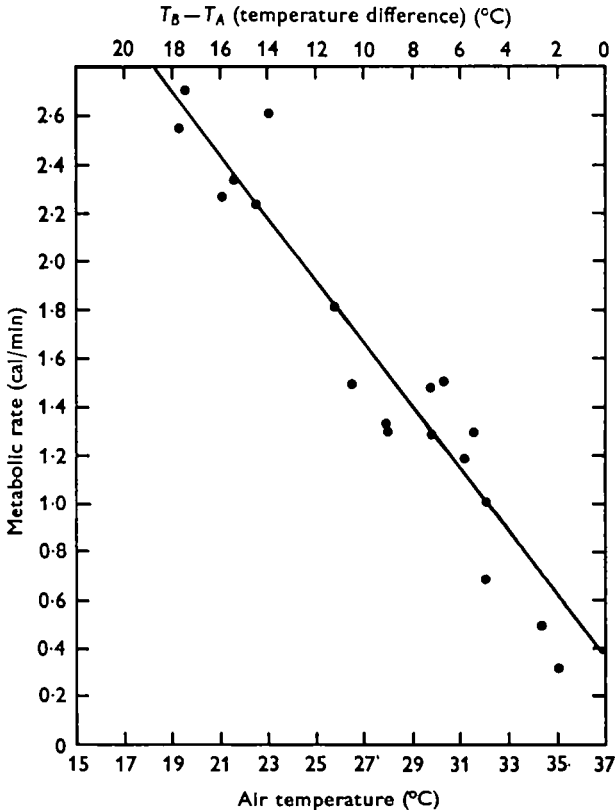
Text-fig. 4. Metabolism of active (flying) moths with respect to the ambient temperature and the temperature difference (T_B-T_A) established during flight. The solid line was drawn by the method of least squares.

Table 2. *Effect of ambient temperature on the measured metabolic rate during activity (flight) and torpor in* Hyalophora cecropia

Sex, air temp. (°C)	Active moth $[M_{AD}]$ (cal/min)	Torpid moth $[M_T]$ (cal/min)	Percentage difference $\dfrac{M_{AD}-M_{AC}}{M_{AD}} \times 100$	Percentage difference $\dfrac{M_{AD}-(M_{AC}+M_T)}{M_{AD}} \times 100$
Male (18)	2·81	0·12	5·3	0·1
Male (25)	1·92	0·21	33·8	25·3
Female (17)	2·93	0·12	7·4	0·0
Female (25)	1·91	0·21	25·4	16·0
Female (30)	1·26	0·30	40·0	20·8

The behaviour of the moths bears out this prediction. Flight activity by all moths is markedly reduced and only a few individuals fly for extended periods. The females generally warm up but do not initiate flight at the peak of the warm-up; rather, they allow the body temperature to fall 3–6 °C. They then initiate a second short warm-up period driving the body temperature back within the range where they can initiate flight. This fluctuation of body temperature by alternate warming and cooling cycles without flight also suggests that the animal does not have a sufficient metabolic capacity for both heat production and flight at these low temperatures.

Table 3. *Stored energy available to* Hyalophora cecropia
during its adult life

Sex	Thoracic wt* $[W_1]$ (g)	Total wt* $[W_2]$ (g)	N	Percentage total wt lipid† (L)	Grams of lipid $(W_2)(L)$	Calories available in lipid stores $[TC] = \dfrac{(W_2)(L)}{9500 \text{ cal/g}}$
Males	0·60±0·15	1·44±0·34	17	33·59	0·484	4598
Females	0·67±0·21	2·68±1·21	41	9·41	0·252	2394

* Mean ±1 standard deviation.
† From Domroese & Gilbert, 1964.

Total available calories. Domroese & Gilbert (1964) found that lipid was the available as well as the preferred substrate for flight muscle in *H. cecropia*. The total lipid content was 33·54 % fresh weight in males and 9·41 % fresh weight in females. Other metabolic reserves such as glycogen and non-glycogen carbohydrates account for less than 1 % of the fresh weight in both males and females.

Table 3 shows the calculated caloric value of the lipid stores of male and female moths based upon the caloric equivalent of lipids as 9500 calories per gram. The mean thoracic weight (W_1) and mean body weight (W_2) are listed. The mean grams of lipid per animal (L_0) was calculated by multiplying the percentage body weight lipid (L) by the total body weight (W_2). The energy pool available to moths in calories (TC) was calculated by dividing the grams lipid (L_0) by 9500 cal/g, the caloric equivalent of lipids. The percentage hydration of lipids was not taken into account in these calculations.

Time of activity. By monitoring thoracic temperature the precise time of activity can be determined. Plate I, shows records of ten males held at 18 °C air temperature. The photoperiod was controlled by an interval timer with a cycle of 16 h light and 8 h dark. Males initiate activity when the lights go off and again when the lights go on. Each light change leads to approximately 1 h of activity. The light period corresponds to the time of sunrise and sunset in June, at this latitude (40° 40′ N), which is the time of emergence of the adult moth from the pupal stage.

Text-fig. 5 shows that the timing of the active periods in male moths is not affected by differences in air-temperature. The pattern of activity in females (Text-fig. 6) differs from that in males. The females become active at the time the lights go off ('sunset') and then gradually decline in activity through the dark period. They do not become active again at the time the lights come on as do males. Again, temperature has no effect on the timing of activity.

The nature of the activity rhythm has not been investigated, but it appears to be

endogenous and not due entirely to changes in light intensity. In the males held at 18 °C the dark period was extended to 6.00 a.m. on the third day of the experiment. The animals initiated activity at 5.00 a.m. as they had on the past 2 days, even though the lights had not turned on. Nevertheless, they also had a second burst of activity at 6.00 a.m. when the lights did come on, which suggests an inherent rhythm in which changes of light intensity is the 'zeitgeber'.
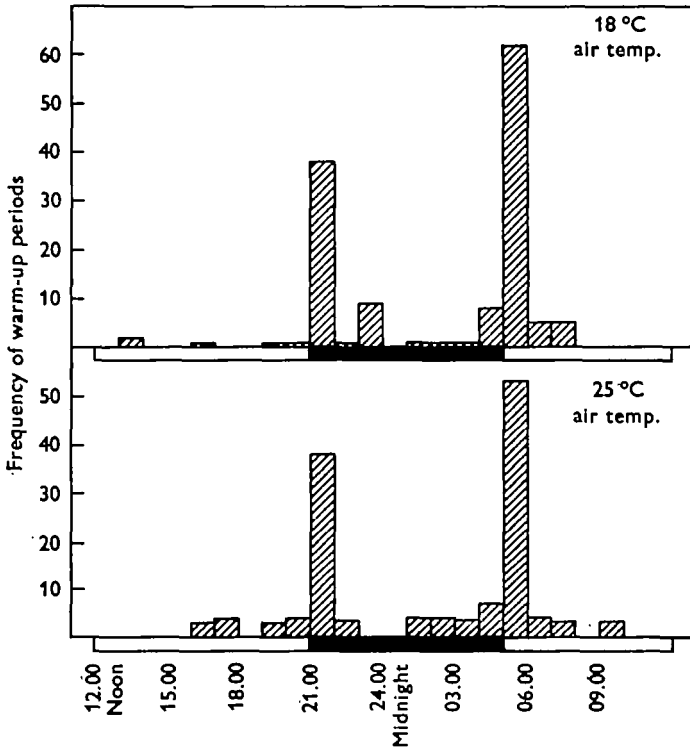
Text-fig. 5. Histograms at 18 and 25 °C air temperature of the frequency of warm-up periods in male moths with respect to time of day and photoperiod.

Number and duration of active (flight) periods. The duration of an activity period is taken as the time of initiation of warm-up to the time at which the animal ceases activity and the thoracic temperature falls to ambient (Text-fig. 1). Table 4 is a summary of the number and duration of active periods. The total number of active periods, (P_A), increases with increasing air temperature in both males and females. There is no significant difference in either the mean life span, (S), between males and females $(P = 0.05)$ or the number of active periods per day (P_D). However, the males are active for significantly longer periods than females. The mean duration of an active period is designated by D_M. The longer active periods, (D_M), of the males is also reflected in the means of the total duration of activity, (D_A). The males are active approximately twice as long as females during their life. The mean duration of torpor, (D_T), represents the difference between the mean life span, (S), and the mean total duration of activity, (D_A).

Table 4. *Effect of temperature on the number and duration of active (flight) and torpid periods in* Hyalophora cecropia

Sex, air temp. (°C)	Number active periods* per animal [P_A]	Mean adult live span [S] days	Active periods* per day [P_D]	N	Duration active periods [D_M] (min)	N	Mean total duration of active periods [D_A] = (D_M)(P_A) (min)	Mean total Duration of torpor [D_T] = (S)−(D_A)
Males (18)	14·33 ± 6·40	7·1	2·10 ± 0·21	9	14·8 ± 11·4	71	212·0	166·9
Males (25)	19·00 ± 5·55	8·1	2·87 ± 0·37	7	22·6 ± 14·2	38	429·4	187·2
Females (17)	12·50 ± 6·40	6·3	2·21 ± 0·75	12	8·0 ± 3·2	64	100·0	149·5
Females (25)	14·70 ± 6·85	6·9	2·91 ± 0·82	9	14·9 ± 10·8	66	219·0	161·9
Females (30)	19·40 ± 6·22	5·9	3·35 ± 1·05	15	19·0 ± 10·9	72	368·6	135·5

* Mean ± 1 standard deviation.

Table 5. *The effect of ambient temperature on the energy balance of* Hyalophora cecropia *during adult life*

Sex, air temp. (°C)	Calculated calories expended, active moths [(M_{AO})(D_A)]	Measured calories expended, active moths [(M_{AD})(D_A)]	Mean cal/h torpid moths [M_{TH}]	Calories expended in torpor [(D_T)(M_{TH})]	Calculated total calories expended [M_T] = (M_{AC}) ×(D_A)+(M_{TB})(D_T)	Measured total calories expended [M_I] = (M_{AD}) ×(D_A)+(M_{TB})(D_T)	Calories available in lipid stores [TC]
Males (18)	563·9	595·9	7·2	1201·5	1765·4	1797·4	4598
Males (25)	724·5	824·4	12·6	2359·2	3083·7	3183·6	4598
Females (17)	272·0	293·0	7·2	1076·6	1348·6	1369·6	2394
Females (25)	313·2	418·3	12·6	2040·6	2353·8	2458·9	2394
Females (30)	280·1	464·4	18·0	2438·3	2718·4	2902·7	2394

Total metabolic expenditure. The total metabolic expenditure of the moths was computed in two ways. The first, or direct method, was to sum the mean total caloric expenditure during activity, (M_{AD}) (D_A), and during torpor, (M_{TH}) (D_{TH}). The sum is expressed as M_X in Table 5. Both the values used to calculate M_X are based on directly measured rates of O_2 consumption which were converted to calories per
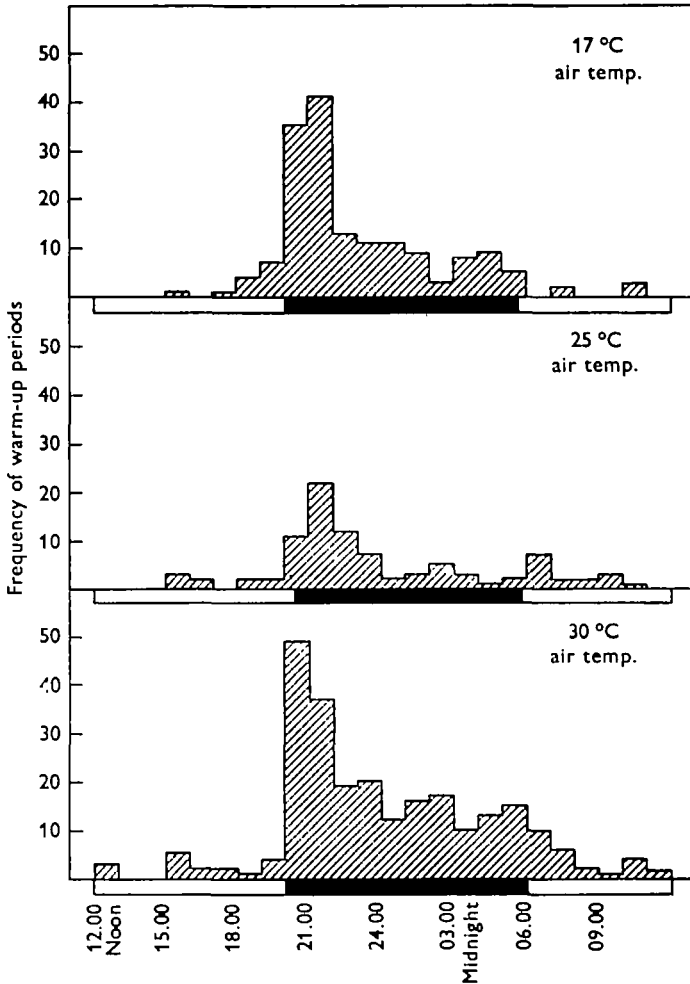
Text-fig. 6. Histograms at 17, 25 and 30 °C air temperature of the frequency of warm-up periods in female moths with respect to time of day and photoperiod.

minute. The second method was to sum the total metabolic expenditure during activity which was calculated from the cooling curves, (M_{AC}) (D_A), and the directly measured O_2 consumption during torpor, (M_{TH}) (D_{TH}), and is expressed as M_Y in Table 5. The function (M_{TH}) (D_{TH}), for torpid metabolism, must be used in both estimates because body temperature equals air temperature during torpor and an estimate of resting metabolism cannot be made from the cooling curve data.

The estimates, M_X and M_Y, for total caloric expenditure of females at 30 °C and the estimate M_Y for females at 25 °C air temperature exceed the total available calories

from lipid stores (*TC*, calculated in Table 3). In all other cases M_X and M_Y do not exceed the total calories available, *TC*. It is significant that in both males and females held at 17–18 °C both estimates of the total metabolic expenditures, M_X and M_Y, fall well below the total calories available (*TC*). This indicates that these animals may die while there are still energy stores available and that it is not lack of substrate that causes death. It is possible that they die from desiccation. Since the animals maintain a mean body temperature of 35 °C when active the saturation deficit may be sufficient to cause a higher rate of water loss in these animals than those held at higher air temperatures.

Flight speed. An estimate of the flight speed was made to determine the distance which both males and females can cover daily and during their entire life. The duration of flight periods was obtained from the body-temperature measurements (Table 4). The flight speed was determined by timing and measuring the distance covered during short flights. Two males and three females were stimulated to warm up and fly by pinching the abdomen. The warm-up period lasts about 3 min at 25 °C air temperature and it was necessary in several observations to stimulate the animal frequently so that it would complete the warm-up process and initiate flight. The observations were made during the day when the animals are not normally active and the duration of flight was very short averaging less than 7 sec. A total of nineteen observations were made on the five individuals. The mean flight speed was 1·88 m/sec, the standard deviation = 0·41, and the range was 1·41–3·00 m/sec.

DISCUSSION

H. cecropia regulates its body temperature within a narrow range when voluntarily active (Hanegan & Heath, 1970). These moths regulate the body temperature physiologically by altering the metabolic rate to offset heat loss depending on air temperature, and also by behavioural mechanisms. The metabolic rate increases with decreasing air temperature, and therefore in an energetic analysis of these insects it is extremely important to account for changes in metabolism corresponding to changes in air temperature.

The calculated metabolic rates for flying moths, (M_{AC}), derived from an analysis of cooling curves are in close agreement with the directly measured rates (M_{AD}). M_{AC} was corrected for convective heat loss which occurs when the animals fly. The measured values, M_{AD}, exceed M_{AC} because the values for M_{AC} do not include the metabolism devoted to the maintenance of the animal, the mechanical output of flight, or the increasing rate of metabolism during the warm-up period.

If the resting metabolism, M_T, is added to M_{AC} the deviation from M_{AD} is reduced (Table 2). However, in both males and females at 18 and 17 °C air temperature the two estimates agree. This predicts that all of the metabolism of an average moth when active is spent in heat production and maintenance, and that there is no reserve for flight at these air temperatures. This is particularly evident in females which in most cases warm up and cool off without initiating flight. Most of the males in these experiments do fly at 18 °C air temperature. The percentage difference between M_{AC} and M_{AD} at 18 °C is 0·1 % and is based on the means of nine individuals (Table 2). In some animals the percentage difference between M_{AC} and M_{AD} is much greater than

0·1%. The variability between individuals tends to reduce the mean percentage difference to the low value of 0·1%.

The mouth parts of the adult are vestigial and it does not feed. The immediate source of energy is the larval fat body that remains during the adult stage. The fresh body weight (W_2) was multiplied by the percentage W_2 lipid (L) to determine the energy reserve of the moth. Lipid as percentage body weight in *H. cecropia* was determined by Domroese & Gilbert (1964). It is evident from Table 3 that the males are significantly smaller in body weight than females yet have approximately twice the energy reserve (TC). This results from the sexual dimorphism in the amount of fat and the metabolically inert eggs carried by the female.

Table 6. *Effect of ambient temperature on the percentage of energy spent in activity by* Hyalophora cecropia

Sex air temp. (°C)	Percentage per adult life $\dfrac{(M_{AD})(D_A)}{M_X} \times 100$	Percentage per day[*]
Males (18)	33·15	34·49
Males (25)	25·89	31·68
Females (17)	21·72	24·11
Females (25)	17·01	23·23
Females (30)	15·99	17·17

[*] Percentage energy spent in activity on a daily basis was calculated as:

$$\frac{(M_{AD})(D_M)(P_D)}{(M_{AD})(D_M)(P_D)+(M_{TB})\,[24-(D_M)(P_D)/60]} \times 100.$$

When the energy reserve is exhausted death occurs. The mean total calories (M_X or M_Y) expended over the life of males and females at 25 and 30 °C air temperature are in close agreement with the calculated energy reserve (TC). Animals at 17–18 °C air temperature may die while there are still energy reserves. The life span at these lower temperatures is not significantly extended as would be predicted from the lower rate of resting metabolism. These animals may die from desiccation. The saturation deficit at a body temperature of 35 °C when the moths are active and an air temperature of 17–18 °C may be sufficient to account for a higher rate of water loss than in animals kept at 25 and 30 °C. When the animals are kept at 5 °C the life span is considerably extended, up to 4 weeks. However, at this low temperature they do not warm up.

Sotavalta & Lavlajainen (1961) were unable to force exhaustion of the flight energy of the moth *Phytometra (Plusia) gamma*. They argued that since the immediate source of flight energy was lipid (Zebe, 1954), flight could be maintained as long as the fat body remained. In the present experiments flight was not forced but voluntary and was initiated by the animals on a rhythmic basis. The percentage of the total metabolic output spent in activity, $[(M_{AD})(D_A)]/M_X \times 100$, was calculated per life and per day (Table 6). Males expend more energy in activity than females. The percentage of energy spent in activity declines with increasing air temperature. This is due to the non-linear increase in resting metabolism (M_T) with air temperature, Q_{10} of 2·12.

It appears that a specific portion of the fat reserve is mobilized each day for activity

and when this is expended the animal becomes inactive. Accordingly both the number of active periods per day (P_D) and the duration of active periods (D_M) increase with increasing air temperature (Table 4). At low air temperatures the temperature difference, T_B-T_A, is greater than at higher air temperatures and it is more expensive for the animal to be active. Since the total calculated calories spent in activity (M_Y in Table 5) remain relatively constant independent of air temperature the moth can afford fewer and shorter active periods at low air temperatures. This suggests metering of the fat reserves for activity on a daily basis. Perhaps the mobilization of fat is controlled by a neural hormone and that either the release or synthesis of this agent has a daily rhythm. Such a neural hormone may in fact serve a dual function: mobilization of a portion of the fat reserve for activity and regulation of the timing of activity.
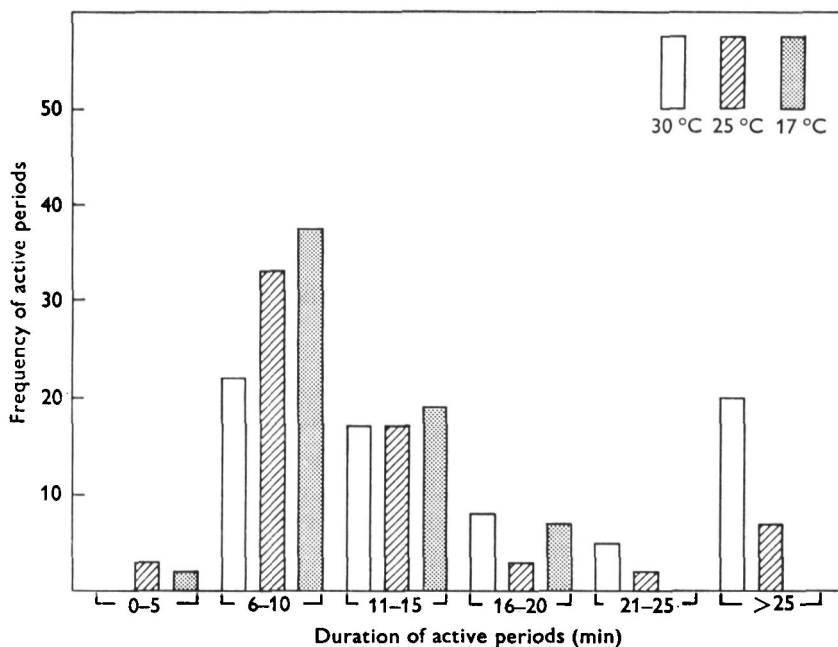
Text-fig. 7. Frequency histogram of the duration of active periods in female moths at 17, 25 and 30 °C air temperature.

The daily rhythm of activity differs in males and females. The males have two periods of activity each lasting approximately 1 h and corresponding to the periods following sunset and dawn in nature. The females have a peak of activity following sunset and the frequency of active periods decreases through the night. Females, unlike the males, lack a second activity period at dawn.

A period of dawn activity in male moths is well known (Rau, 1916; Sweadner, 1937; and others), however, a period of activity following sunset has been previously reported to occur only in rare instances (Sweadner, 1937). If the female releases the sex-attracting pheromone during and following the peak of her activity, which corresponds to the first activity period of the male, it could serve as a general stimulus for concentrating males in the vicinity of the virgin female. During this period the males would fly into the wind following the 'pheromone trail' emitted by the female. Males

may rarely encounter the female at this time and begin copulation (Sweadner, 1937). They probably do not contact the female at this time but are close proximity to her. Approximately 1 h after sunset both males and females become inactive. At dawn, the females are inactive, but they may continue to release or have a second period of release of the pheromone. The males may locate the females through both visual and olfactory stimuli during their dawn activity period.

The males are most susceptible to predation when they make their long-range evening flights. The primary predators of moths are birds, which are generally inactive after sunset. Therefore, by making the long-range flight after sunset the males may escape heavy predation by birds. At dawn when the male actually locates the female the predation by birds would be very high. However, at this time the flight range of the male is very short, thereby reducing their exposure.

Copulation normally occurs just after dawn (Rau, 1911) and normally lasts until the following evening. Sweadner (1937) reported that although copulation lasts for 12 or more hours only 2 h are actually needed for insemination. The long duration of copulation is probably due to the inactivity of both males and females during the daylight hours. At sundown the female becomes active, dislodges the male, and begins her egg-laying flights which may last through the night.

The females do not have to be fertile to begin egg-laying. In this study all females were virgin, yet they began egg deposition on the third to fourth day after eclosion. They normally lay eggs in clusters of three to six. The thoracic temperature measurements show a cycling of warm-up followed by flight and then a short period of cooling, which corresponds to the period of egg deposition. This distinct cycling of thoracic temperature occurs during the night and may last up to $1 \cdot 5$ h.

The mean duration of active periods (D_M) increases at higher air temperatures. Although D_M was used to calculate the metabolic expenditures of females it appears that the duration of active periods increases on the day before death. Text-fig. 7 shows the frequency of active periods with respect to duration. It can be seen that there is a bimodal distribution in the females held at 25 and 30 °C air temperature. The females held at 17 °C do not exhibit the long active periods. At this low air temperature females probably do not have the metabolic reserves to maintain body temperature at the level required for flight.

A long active period may reflect a reduced flight load because the female has deposited most of her eggs. During the time after copulation when the active periods are short the flight range would be limited. This would restrict egg deposition to the locality in which the female developed and survived, thereby increasing the chances of survival of the larva. A long flight at the end of a female's life may function to disperse eggs to new localities.

Male moths are capable of flying long distances. Rau (1916) reported that 40% of tagged males returned to caged females from a distance of one-eighth mile (km) and 11% returned from a distance of 3 miles (km). In one case a tagged male returned to a caged female from a distance of 5·5 miles (Weast, 1959). This flight took 3 days and the experiment was performed in the Arizona desert where no pheromones from other saturniid species could interfere with the flight of the male. Weast also reported that females of *H. promethea* attract males over a distance of 14 miles (23·3 km) from Milwaukee to Waukesha, Wisconsin. *H. cecropia* may have a similar flight range.

Table 7. *Calculated range of Hyalophora cecropia at 25 °C ambient temperature*

Range per day

Sex	Flight speed [V] (m/sec)	Duration of* activity $[D_M]$ (min)	Maximum duration of activity $[D_M + 1\ \text{s.d.}]$ (min)	Number of active periods $[P_D]$	Flight range $(V)(D_M)(P_1)$ (km)	Maximum flight range $(V)(D_M + 1\ \text{s.d.})(P_D)$ (km)
Male	1·88	22·6 ± 14·2	36·8	2·87	7·3	11·9
Female	1·88	14·9 ± 10·8	25·7	2·91	4·9	8·4

Range per adult life

	Total active periods $[P_A]$	Flight range $(V)(D_M)(P_A)$	Maximum flight range $(V)(D_M + 1\ \text{s.d.}) \times (P_A)$
Male	19·0	48·4	78·7
Female	14·7	24·7	42·6

* Mean ± 1 standard deviation.

Table 7 lists the range of both males and females at 25 °C air temperature calculated from the duration of activity (D_M) and the mean flight speed.

The ranges listed are for straight-line flights. The male tracks the female through detection of the wind-borne pheromone. Allowing only one-third of the calculated distances for deviations from a straight-line course the mean distance covered by males per day would be 2·4 km and a maximum of 4·0 km. This is certainly sufficient to account for the distance covered by males reported by Rau (1916). Over a period of 3 days the mean distance covered would be 7·2 km with a maximum distance of 12·0 km, which would easily account for the 5·5 mile (9·1 km) flight reported by Weast (1959).

Although populations of *H. cecropia* appear to be isolated by habitat restrictions there may be considerable contact between these apparently isolated populations because of the long flights of the males.

SUMMARY

1. The time of activity and the duration of active periods (flight) of moths of the species *Hyalophora cecropia* has been determined by monitoring thoracic temperature.

2. The metabolic cost of flight per day and per adult life has been determined directly by measuring O_2 consumption and indirectly by analysis of cooling curves of individual moths.

3. An energy balance sheet has been derived which gives the metabolic cost of flight and maintenance (during torpor) over the insect's adult life.

4. The metabolic stores mobilized for daily activity appear to be fixed and independent of air temperature. This mobilization of fat stores may be under hormonal control.

5. It is metabolically more expensive for moths to be active at low air temperatures. The number and duration of active periods at low air temperatures is reduced, but, the metabolic expenditure for activity is equal to that of animals held at higher air temperatures.

6. Females have a smaller total energy reserve than males. The number of active periods per day is not significantly different between the sexes at any given temperature, but in females the active periods are significantly shorter in duration.

7. The flight speed has been determined, and estimates of the flight range per day and per adult life have been calculated.

8. The ecology of *H. cecropia* has been discussed with respect to the timing and duration of active periods, the range and speed of flight, and the energetic cost of flight and maintenance metabolism.
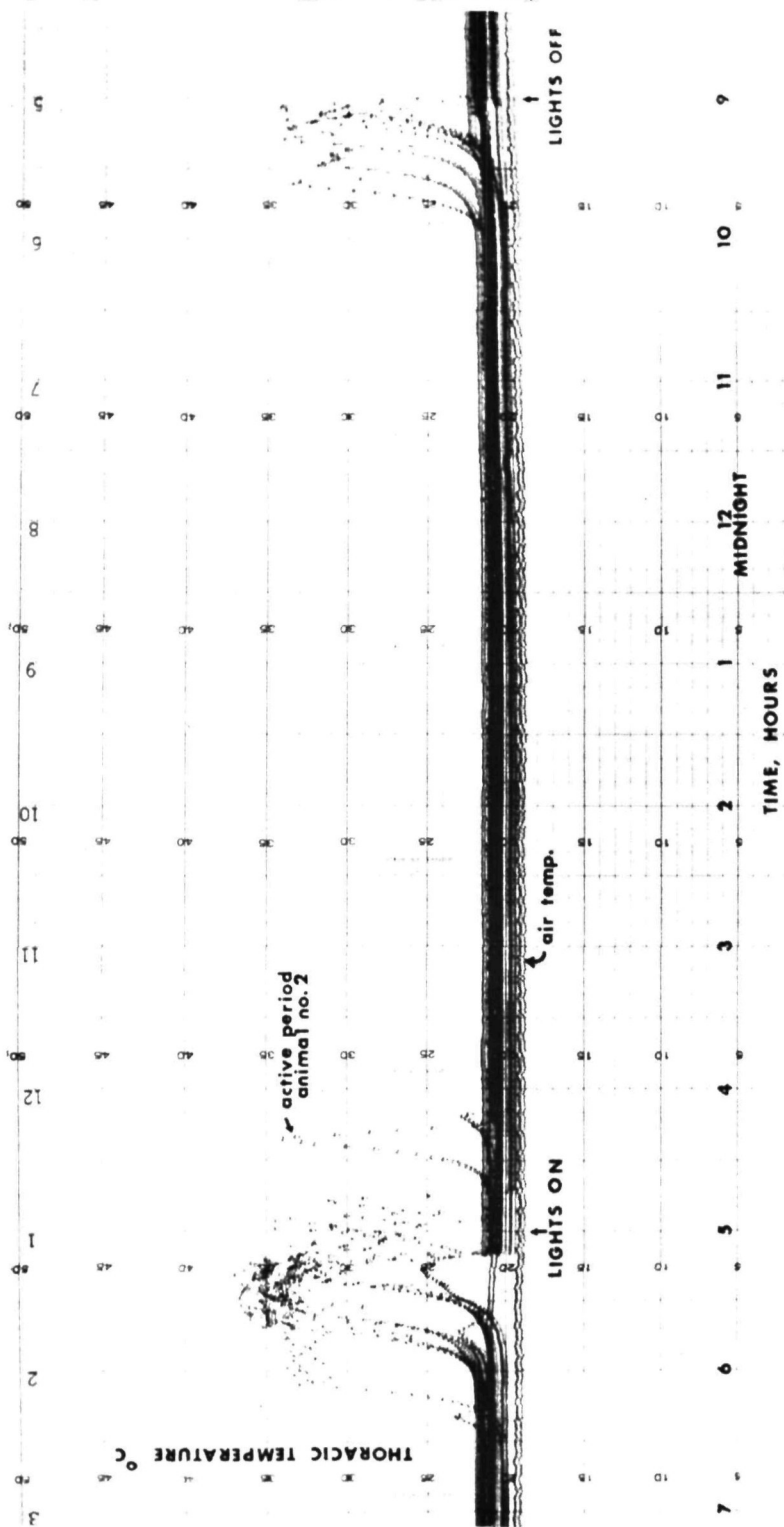
REFERENCES

DOMROESE, K. A. & GILBERT, L. I. (1964). The role of lipid in adult development and flight metabolism in *Hyalophora cecropia*. *J. exp. Biol.* **41**, 573–90.

HANEGAN, J. L. & HEATH, J. E. (1970). Mechanisms for the control of body temperature in the moth, *Hyalophora cecropia*. *J. exp. Biol.* (In the Press.)

HEATH, J. E. & ADAMS, P. A. (1969). Temperature regulation and heat production in insects. In *Experiments in Physiology and Biochemistry*, Vol. 2, Ed. G. A. Kerkut. New York: Academic Press.

KROGH, A. & ZEUTHEN, E. (1941). The mechanism of flight preparation in some insects. *J. exp. Biol.* **18**, 1–10.

RAU, P. (1910). Observations on the duration of life, on copulation and oviposition in *Samia cecropia*, Linn. *Trans. Acad. Sci. St Louis* **19**, 21–48.

RAU, P. (1911). Further observations on copulation and oviposition in *Samia cecropia* Linn. *Trans. Acad. Sci. St Louis* **20**, 309–19.

RAU, P. & RAU, N. (1916). The sex attraction and rhythmic periodicity in the giant saturniid moths. *Trans. Acad. Sci. St Louis* **26**, 83–221.

SOTAVALTA, O. & LAULAJAINEN, E. (1961). On the sugar consumption of the drone fly (*Eristalis tenax L.*) in flight experiments. *Anns Acad. Sci. Fennicae* A **453**, 1–25.

SWEADNER, W. R. (1937). Hybridization and the phylogeny of the genus platysamia. *Ann. Carnegie Mus.* **25**, 163–242.

WEAST, R. D. (1959). Isolation mechanisms in populations of *Hyalophora* (Saturniidae). *J. Lepidopt. Soc.* **13**, 213–16.

WEIS-FOGH, T. (1952). Fat combustion and metabolic rate of flying locusts (*Schistocerca gregaria* Forskål). *Phil. Trans.* B **237**, 1–36.

ZEBE, E. (1954). Uber den Stoffwechsel der Lepidopteren. *Z. vergl. Physiol.* **36**, 290–317.

EXPLANATION OF PLATE

Thoracic temperature measurements of 10 male moths at 18 °C air temperature, showing the timing of activity with respect to photoperiod.