of these equations, but these data are simply not sufficient for indicating which, if any, of the three possible models is valid.

While we could reject the linear model in this case as it gives incorrect dates for the divergence of man from the African apes, this is not the case for the other two models. One model agrees with conventional estimates for the primate time scale and the other, while giving debatable results, cannot be rejected out of hand. We thus conclude that no one of these models without better evidence can be assumed valid. Mathematical models of this type must be considered questionable if based on such scanty data.

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8. G. G. Simpson, personal communication, as quoted in A. S. Hafleigh and C. A. Williams, Jr. (*2*).
9. The variation in ID values tends to increase as the phylogenetic distance increases. The data from Wilson *et al.* (*6*) for the bullfrog indicates a range of 14 to 40 ($s^2 = 135.0$). Sarich's data (*4*) is also variable, with a range of 8.6 to 18.0 for the Prosimii ($s^2 = 12.4$).
10. Our ID value for the duck, pigeon, and ostrich, obtained from Wilson *et al.*, differs from the value indicated by Sarich (*4*). However, the discrepancy is minor.
11. In each case, s^2 was computed from observed ID values and predicted ID values derived from each of the equations.
12. S. N. Salthe and N. O. Kaplan, *Evolution* **20**, 607 (1966).
13. The power function provides a perfect fit to two points since two points plotted on log paper can be fitted exactly by a straight line.
14. Sarich has opted for a more recent divergence (30 million years) (*5*) than generally accepted, partly on the basis of scanty fossil data. Simpson, however, representing the viewpoint of a relatively early divergence, has suggested a range of 35-45 million years (*8*).
15. Sarich bases his 3.5 million and 5 million years for the divergence of man from the African apes on a single point in each case (*4, 5*), whereas the proper statistical procedure is to fit a regression line to both of these points (and going through the origin), which gives a date of 3.8 million years for this divergence.
16. We thank D. R. Ackerman, B. J. Williams, and P. S. Miller for review of the manuscript.

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Thoracic Temperature Stabilization by Blood Circulation in a Free-Flying Moth

Abstract. *The sphinx moth,* Manduca sexta, *maintains its thoracic temperature within a degree of 42°C while in free flight over a range of air temperatures from about 17° to 32°C. Tying off the dorsal vessel abolishes temperature control. Moths with tied off vessels overheat and then stop flying at air temperatures of about 23°C. However, flight at this temperature is possible when the thoracic scales are removed. The mechanism of temperature control involves transfer of the heat produced in the thorax to the blood pumped from the dorsal vessel, and the subsequent dissipation of this heat when the blood returns to the relatively cool abdomen.*

Most members of the family Sphingidae are fast-flying, narrow-winged, nocturnal moths which feed on nectar while they hover. They range in weight from about 200 mg to 4 g and occur from the equator to boreal regions.

The adults of the tobacco hornworm *Manduca sexta* used in this study weighed from 1.8 to 2.6 g, which is in the same range as the smallest hummingbirds. Like other sphingids, *M. sexta* undergoes a warm-up involving "wing-whirring" before flight (*1*), a process analogous to shivering that results in the elevation of thoracic temperature by the production of endogenous heat. Flight is not initiated until thoracic temperature reaches approximately 38°C. High thoracic temperature is apparently necessary for controlled hovering, which may require a nearly "maximum" effort (*2*). At low ambient temperature the heat produced during flight is readily lost to the environment, but at high ambient temperatures this endogenous heat can result in excessive elevation of thoracic temperature unless rates of heat loss are increased. I now report that circulation of blood affords a mechanism for the control of temperature in the thorax during free flight at a variety of ambient temperatures.

The laboratory population used in this study was reared from eggs obtained in the western Mojave desert between Lancaster and China Lake, California, where the animals are active from early June to September. The larvae were reared on tobacco leaves through the 4th larval instar, and on a prepared diet of wheat germ, agar, and additives (*3*) during the last larval instar.

Thoracic temperatures were obtained from moths that had undergone warm-up and had flown for 2 minutes in a controlled-temperature room with an interior volume of 5.0 m³. Thoracic temperatures were measured by inserting a sharpened, 30-gauge copper-constantan thermocouple into the center of the thorax, and they were registered on a recording potentiometer within 5 seconds of the time of capture of the flying moth. Unless otherwise specified, each moth was measured only once.

After 2 minutes of flight, mean thoracic temperatures were between 41° and 42°C over a range of ambient temperatures from 20° to 30°C (Fig. 1). Below 20°C thoracic temperatures were lower, and at 12.5°C they averaged 37.7°C. At 12°C or less many moths did not arouse when stimulated. At an ambient temperature of 35°C, moths often had to be prodded to keep them flying, and thoracic temperatures increased to a mean of 43.3°C. The moths were reluctant to fly continuously at ambient temperatures above 35°C, although they could fly for about 1 minute even at 41°C.

If the amount of work performed during flight is independent of ambient temperatures, these data suggest that at ambient temperatures above 20°C the moths can increase the rate of heat loss so that thoracic temperature remains essentially uniform.

The temperatures toward the posterior end of the abdomen of *Manduca sexta* during flight were only slightly above ambient. Thus the abdomen is a potential site of heat loss if the heat produced in the thorax during flight can be transferred to it by the circulatory system. The dorsal vessel is well developed in sphingids and is visible through the cuticle in all seg-

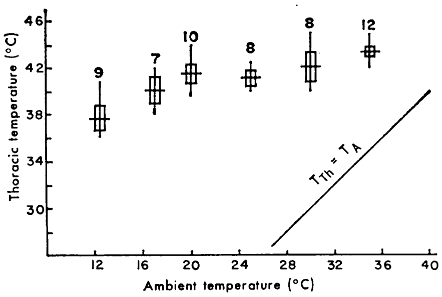

Fig. 1. Thoracic temperature T_{Th} during flight plotted against ambient temperature (T_A) of 54 moths. At each T_A the vertical line represents the range, the horizontal line represents the mean (*m*), and the boxes enclose the interval $m \pm 2$ standard errors.

ments of the abdomen if the scales are removed (Fig. 2). Pulsations of the dorsal vessel travel from posterior to anterior. Observations of over 25 moths indicate that immediately after flight at ambient temperatures above 30°C two to three pulses per second pass along the dorsal vessel carrying blood into the thorax, but below 20°C no such pulsations were observed. Between 20° to 30°C pulsation was irregular and often restricted to that portion of the vessel in the anterior part of the abdomen.

To determine whether or not the blood pumped from the cool abdomen into the relatively hot thorax actually results in significant reductions of thoracic temperature, the dorsal vessel was ligated with surgical thread in the first segment of the abdomen. Moths with the dorsal vessel tied off, but otherwise intact, were flown for 2 minutes over a range of ambient temperatures from 15° to 23°C. The thoracic temperature of these moths increased directly with ambient temperature and the temperature difference averaged 23°C (Fig. 3). At ambient temperatures above 23°C few of the moths could be induced to continue flying for the full 2 minutes (most moths with unligated vessels could fly for as long as 2 minutes even at 35°C or above). These data are consistent with the hypothesis that circulating hemolymph serves in the dissipation of heat from the thorax to the abdomen during flight at high ambient temperatures. However, it does not exclude the possibility that ligation affected some aspects of the flight machinery other than heat loss.

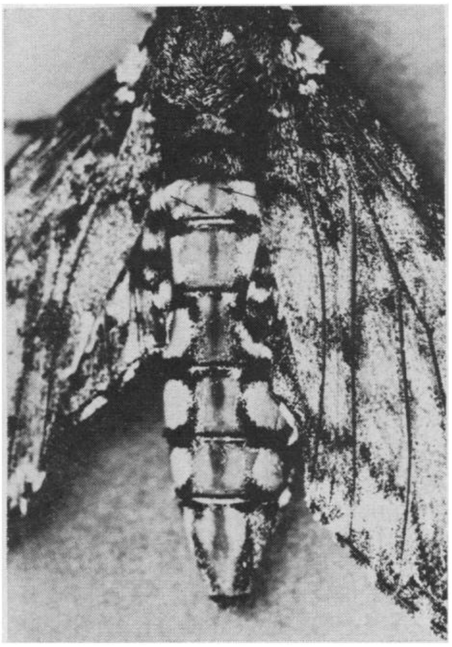

Fig. 2. Dorsal view of abdomen with scales partially removed. The dorsal vessel was ligated at the arrows.

Manduca sexta is covered with a thick (2 mm) layer of scales on the thorax and a thin (0.5 mm) layer on the abdomen. These scales have an insulating function (*4*). The removal of scales should, therefore, increase thermal conductance and thus allow one to differentiate between the effects of ligation on heat loss and its possible effect on other aspects of the flight machinery.

The thoracic scales were removed from a series of moths, and their dorsal vessels were ligated. They were flown for 2 minutes at ambient temperatures from 21° to 30°C. Their thoracic temperatures averaged about 15°C above ambient (instead of 23°C when scales were present) and again paralleled ambient closely (Fig. 3). These moths were able to fly for 2 minutes at ambient temperatures as much as 7° or 8°C higher than those similarly ligated but with their scales present. Thus, ligation of the dorsal vessel inhibited flight at ambient temperatures in excess of 23°C by destroying the circulatory machinery which was used for heat dissipation.

The sphinx moth *Celerio lineata* may have some control of its thoracic temperature during tethered flight (*5*). The mechanism of control that was suggested for *Celerio* involved control of the rate of heat production (*5, 6, 7*). In contrast, the rate of heat loss in *M. sexta* is varied. Furthermore, normal moths can fly in a wide range of ambient temperature in which they maintain a nearly stable thoracic temperature. At ambient temperatures below 20°C the slopes of the regression lines for thoracic on ambient temperature are significantly different between normal and ligated moths ($P < .05$). This indicates that differences in the rate of heat loss are occurring not only at high but also at low ambient temperature. At ambient temperatures above 20°C the thoracic temperature of normal moths stabilizes, whereas ligated moths begin to overheat. At 23°C the ligated moths stop flying because of overheating (ligated animals with the insulating layer of scales removed will fly at this temperature). It seems probable that normal moths lose heat to the abdomen by way of the circulatory system since they do fly at relatively high ambient temperatures without overheating, whereas moths in which the circulation has been interrupted do not. These results indicate that the blood circulation of an insect may be functional in changing rates of heat loss and in stabilizing thoracic temperature. This mechanism, which may involve increases as well as restrictions of blood flow, allows the sphinx moth *Manduca sexta* to fly in a relatively wide range of ambient temperature.

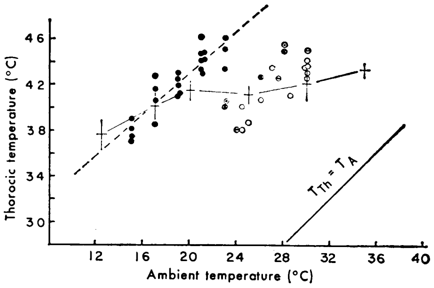

Fig. 3. Thoracic temperatures during flight plotted against T_A in moths with the dorsal vessel ligated. The broken line represents the regression of T_{Th} versus T_A of moths with no scales removed. The 23 animals with scales on the thorax are designated by the shaded circles, while the unshaded circles represent measurements from 14 moths without thoracic scales. Several measurements were taken from some of the latter moths, which are represented by similar symbols. The thin solid line connects the averages of the flight temperatures of normal moths. The vertical bars give the 95 percent confidence intervals.

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DDT Metabolism: Oxidation of the Metabolite 2,2-bis(p-Chlorophenyl)ethanol by Alcohol Dehydrogenase

Abstract. *A metabolite of DDT, 2,2-bis(*p*-chlorophenyl)ethanol, is a substrate of crystalline liver alcohol dehydrogenase. The oxidation of the substrate was detected spectrophotometrically. The* p*-nitrophenylhydrazone derivative of the product, 2,2-bis(*p*-chlorophenyl)acetaldehyde, was identified by comparing its mass spectrum and thin-layer chromatographic behavior with that of an authentic sample.*

The metabolism of DDT [1,1,1-trichloro-2,2-bis(*p*-chlorophenyl)ethane] in mammals gives rise to 2,2-bis(*p*-chlorophenyl)ethanol (DDOH) and 2,2-bis(*p*-chlorophenyl) acetic acid (DDA) (*1*). An intermediate aldehyde has been proposed but has not yet been found to occur in vivo. We have synthesized the proposed intermediate, 2,2-bis(*p*-chlorophenyl)acetaldehyde (DDCHO) (*2*).

Because the aldehyde has never been found in vivo and because preliminary studies indicate that the synthetic compound is highly unstable and reactive, the aldehyde was examined as a possible product of oxidation of DDOH by crystalline liver alcohol dehydrogenase (E.C.1.1.1.1).

The oxidation of DDOH was detected in a double-beam spectrophotometer by following the reduction of nicotinamide-adenine dinucleotide (NAD) at 340 nm in the presence of crystalline horse liver alcohol dehydrogenase (*3*). Because DDOH is insoluble in aqueous media, the compound was dissolved in 50 percent glycerolformal before its addition to the buffered incubation media. The resulting cloudy suspension prevented accurate spectrophotometric determination of reaction rates, but definite increases in absorbance were observed. No reaction was detected in the absence of enzyme or NAD. The reverse reaction, the reduction of DDCHO, was similarly observed with the substitution of reduced NAD for the oxidized form and by following the decrease in absorbance at 340 nm. Glycerolformal in the absence of DDOH also catalyzes the reduction of NAD, but the reverse reaction was not observed for glycerolformal.

Direct chemical evidence for the enzymatic oxidation of DDOH to DDCHO was obtained by formation of the *p*-nitrophenylhydrazone derivative. An incubation mixture was prepared containing 0.016*M* sodium pyrophosphate, *p*H 8.8, 0.008*M* NAD, and 2 mg of DDOH in a final volume of 6 ml. Crystalline liver alcohol dehydrogenase (2 mg) was added, and the mixture was incubated at 37°C for 30 minutes; then 0.01*M* *p*-nitrophenylhydrazine (0.5 ml) was added, and the mixture was shaken. A chloroform extract of the mixture was prepared and evaporated to a minimum volume. Portions of the extract were chromatographed on silica-gel plates with a mixture of benzene and petroleum ether (75:25) or benzene and ethyl acetate (95:5). The extract yielded a spot on the chromatograms whose R_F values, 0.28 and 0.12 for the respective solvent systems, corresponded to those of the authentic *p*-nitrophenylhydrazone derivative of DDCHO.

The identity of the derivative obtained from the enzymatic incubation was established by low-resolution, electron-impact mass spectrometry at 20 ev. High resolution measurements (*4*) confirmed the elemental composition of the ion fragments observed in the low resolution spectrum. An authentic sample of the *p*-nitrophenylhydrazone of DDCHO shows a prominent molecular ion at *m/e* (mass to charge) 399 and the base peak at 261. Other prominent ions of interest were *m/e* 249, 235, 226, 199, 200, 125, 122, and 111. The mass spectrum of the chloroform-extractable derivative was the same as that of the authentic sample of the DDCHO derivative. These findings strongly support the probability that DDCHO is a metabolite of DDT.

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Phenolic Aldehydes: Generation from Fossil Woods and Carbonaceous Sediments by Oxidative Degradation

Abstract. *Aromatic aldehydes derived from fossil woods and carbonaceous sediments were identified by gas-liquid chromatography; their geochemical significance is discussed.*

To help characterize organic matter in sediments and to test the possible significance of phenolic aldehydes as geochemical indices, we have analyzed oxidation products for *p*-hydroxybenzaldehyde, vanillin, and syringaldehyde, three products of mild oxidation of lignin in woody tissues. The samples were ground with a mortar and pestle, Wiley mill (contemporary woods), or Angstrom disk mill (indurated sediments), and sieved to 60 mesh. Oxidation was carried out in a stainless steel tube in an oil bath at 183°C with about 15 ml of 8 percent aqueous NaOH and 1 ml of base-washed, redistilled nitrobenzene per gram of sample, for about 2 hours (*1, 2*). The reaction mixture was filtered, and the filtrate was washed with methylene chloride to remove nonpolar byproducts. The aqueous phase was acidified with 6*N* HCl, and the phenolic aldehydes were extracted with methylene chloride. The extract was concentrated