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EVAPORATIVE COOLING AND RESPIRATORY  
ALKALOSIS IN THE PIGEON\*

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*Communicated February 18, 1966*

Some birds can withstand exposure to high air temperatures and keep their body temperature from rising to a lethal level by increasing evaporation of water. This is achieved by an increase in respiratory ventilation similar to panting in dogs. Most noticeable in birds is a change in respiratory rate which may increase more than 20-fold. An increase of this magnitude should have considerable consequences for the gas exchange in the lungs. In particular, the removal of carbon dioxide can be expected to increase, and unless this loss can somehow be prevented, the result will be a severe alkalosis.

The respiratory system of birds differs from that of mammals in that the lungs communicate with large, membranous air sacs which are thought to act as bellows. The air can reach the air sacs via two different pathways, either directly through the mesobronchi, or via the numerous, smaller parabronchi which penetrate the lung parenchyma (for anatomical details, see refs. 1 and 2). It has been suggested that this arrangement could permit air to pass to and from the air sacs without passing the gas exchange surfaces of the lung.<sup>2-5</sup> Thus, respiratory ventilation could increase without causing alkalosis due to removal of CO<sub>2</sub> from the lungs, but experimental evidence that could clarify this point has been inadequate. We have approached this problem by examining the acid-base balance of the blood, and found that during heat regulation pigeons sustain a severe alkalosis due to excess loss of CO<sub>2</sub>.

*Methods.—Animals:* Adult domestic pigeons (*Columba livia*) weighing 270 to 425 (mean = 347) gm were obtained commercially. They were kept and fed in outside aviaries, but were maintained inside at ordinary room temperature for one week preceding any experiment.

*Temperature measurements:* Air and body (cloacal) temperatures accurate to  $\pm 0.2^\circ\text{C}$  were recorded with copper constantan thermocouples connected to a Leeds and Northrup Speedomax G 16-channel potentiometer. Air temperatures above  $50^\circ\text{C}$  were measured with a mercury thermometer. Cloacal thermocouples were inserted about 2 cm and taped to the tail feathers. All

experiments were conducted in a temperature-controlled room ( $\pm 1^\circ\text{C}$ ) and at a relative humidity of less than 20%.

*Evaporation and oxygen consumption* were determined simultaneously in an open system. The respiration chamber was a sealed 5-gallon tin can, ventilated at 2.4–3.0 liters/min by dry air which had passed over a column of Drierite ( $\text{CaSO}_4$ ). Blank determinations were used to check the dryness of the air. Water was collected from the air leaving the chamber by freezing in glass U-tubes immersed in an alcohol-dry ice bath. Known amounts of water evaporated into the system could be recovered within 2%. At 50–51°C the rate of evaporation was so high that chamber humidity increased enough to affect the birds, and body weight changes in open-wire cages (corrected for weight loss due to droppings and carbon dioxide loss) were used to estimate evaporation rates. Oxygen consumption was measured with a Beckman G-2 recording paramagnetic oxygen analyzer. The measurements were made after 1 hr or more when the oxygen consumption trace indicated that the birds had quieted down.

*Respiratory rates:* At room temperature the respiratory rate was sufficiently low (about 30 cycles/min) to be counted directly. At high temperature the frequency was measured to  $\pm 1\%$  with a stroboscopic tachometer (General Radio Strobotac 631-BL).

*Blood samples* were drawn from polyethylene tubing (Intramedic PE-50) placed in the ulnar artery and filled with heparinized saline (30 USP units/ml). The catheter extended outside the cage so that blood could be withdrawn without handling or immobilizing the bird. Samples (about 0.3 ml) were withdrawn anaerobically and the catheter was flushed with saline between samples. The total amount of blood taken during any experiment was less than 10% of the estimated blood volume. During measurement of respiratory rates and blood sampling, drinking water was available to eliminate any effect of dehydration on panting.

*Partial pressure of  $\text{CO}_2$  ( $P_{\text{CO}_2}$ ) and pH* were measured according to the method of Astrup,<sup>6</sup> using a Radiometer AME-1C micro-pH apparatus. In this method two aliquots of the blood sample are equilibrated with gas mixtures at two different  $\text{CO}_2$  concentrations and the pH of each aliquot is measured. By interpolation, the  $P_{\text{CO}_2}$  of the blood, as drawn directly from the bird, can be estimated from the pH. The temperature of the blood during equilibration and pH measurement was kept within  $\pm 0.5^\circ\text{C}$  of the bird's temperature.

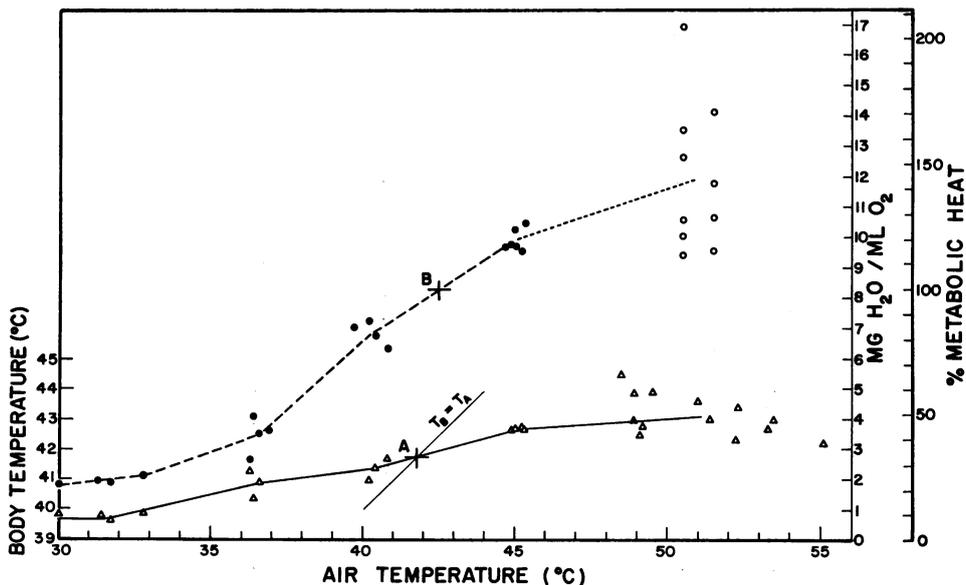


FIG. 1.—Evaporation (broken line) and body temperature (solid line) of pigeons at various air temperatures. Evaporation: open circles above 50°C represent values estimated from weight loss. The cross marked B indicates where 100% of the metabolic heat is dissipated by evaporation. Temperature: each point is the mean for 30 min for one bird. The cross marked A is where body temperature ( $T_B$ ) equals air temperature ( $T_A$ ) (cf. text).

*Tolerance to High Air Temperature.*—The pigeons could tolerate air temperatures of 50°C for several hours. The body temperature would initially rise somewhat and would then stabilize. At all air temperatures above 41.8°C (Fig. 1, point A), the birds maintained their body below air temperature, and at the highest temperatures (mean = 50.9°C) the mean body temperature was  $43.1 \pm 0.7^\circ\text{C}$  s.d. (14 birds). Occasionally a bird became panicky, and the ensuing exertion, which interrupted orderly panting, caused body temperature to rise explosively. The cloacal temperature of one bird reached 46.1°C, and the bird survived after immediate cooling under cold water. Also, in other birds, survival at body temperatures above *ca.* 46°C is limited to a few minutes' exposure.<sup>7</sup> From this it is apparent that the 43.1°C mean recorded for pigeons at 51°C represents regulation well below lethal levels.

The amount of water evaporated increases sharply with increasing air temperature (see Fig. 1, broken line). Up to 37.5°C our figures are in agreement with those previously reported by Kayser.<sup>8</sup> Kayser made no measurements at high temperatures, and later authors concluded that the maximum capacity for evaporative cooling in pigeons had been measured.<sup>9</sup> The extension of our results to temperatures above 50°C shows that the pigeon has an excellent capacity for cooling by evaporation.

At low air temperatures the metabolic heat is dissipated partly by evaporation, but mainly by conduction and radiation. At the point where body temperature equals air temperature, there is no heat flow between environment and the body except by evaporation which must equal the heat produced by metabolism. In other words, at this point 100% of the metabolic heat produced is lost by evaporation (see Fig. 1, scale at far right). Above this point (42.5°C, marked B on broken line) the further increase in evaporation can be used to estimate heat flow from the hot environment to the bird. Ideally, points A and B should occur at the same temperature, and the close agreement of the evaporation data (point B) with the temperature measurements (point A) indicates the accuracy of the metabolic and evaporation measurements.

*Respiratory Rates and Blood  $P_{\text{CO}_2}$  Changes during Panting.*—Pigeons exposed to high temperatures panted at rates more than 20 times the breathing rate at moderate temperatures. The panting consisted of thoraco-abdominal movements and synchronous fluttering of the gular region. The mean respiratory rate at 23–40°C was  $29 \pm 3/\text{min}$  (s.d.), while at 50°C it was  $650 \pm 60$ . The transition from regular breathing to panting is not gradual; at moderate heat load the bird pants intermittently, alternating between periods of regular breathing, with rates one to two times the normal, and periods of full panting. The same pattern is observed in the dog.

In mammals hyperventilation causes loss of  $\text{CO}_2$  from the blood and alkalosis. Contrary to the assumption that an increased respiratory volume in birds need not involve the gas exchange in the lung, our measurements of blood  $P_{\text{CO}_2}$  and pH during panting in the pigeons (Fig. 2) indicate considerable involvement.

Curve I represents the relationship between  $P_{\text{CO}_2}$  and pH for samples from resting birds at 21.5–26°C, and point A gives the mean value and the standard deviation for the pH of the blood. Curve II represents blood samples from the same birds after panting for 14 min to 6 hr (mean 2.1 hr) at 48–54°C. The mean blood

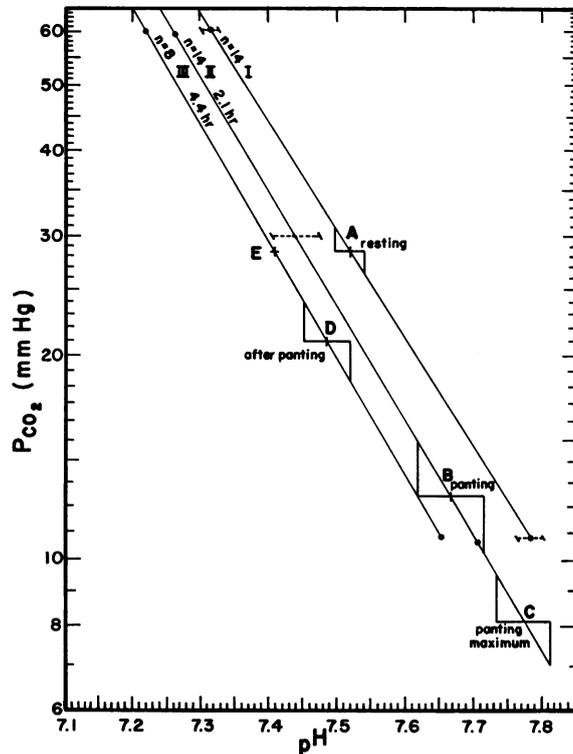


FIG. 2.—Equilibrium curves and mean blood pH and  $P_{CO_2}$  values for nonpanting pigeons (curve *I*), during panting (curve *II*), and immediately following cessation of panting (curve *III*). The standard deviation for curves *I* and *II* is indicated by horizontal dotted lines. For discussion of points *A–E*, see text.

pH had shifted from a nonpanting  $7.52 \pm 0.02$  to  $7.69 \pm 0.05$ , and the corresponding mean  $P_{CO_2}$  had decreased from  $28.5 \pm 2.3$  to  $12.3 \pm 3.3$  mm (point *B*). The preceding means include all determinations, while the mean of the highest pH values recorded for each of the 14 birds was  $pH = 7.79 \pm 0.04$  (point *C*). The corresponding mean minimum  $CO_2$  value was  $8.1 \pm 1.4$  mm. The highest single pH value recorded was  $7.965$ , with  $P_{CO_2} = 6$  mm.

It is interesting to note that curve *II* is shifted to the left as compared to the normal condition (curve *I*). The shift was not due to temperature, because aliquots equilibrated at the two temperatures differed by no more than  $0.02$  pH. The shift was not an artifact since the pH values were measured directly and the simultaneous  $CO_2$  values were obtained by equilibration with known gases. Therefore, it indicates that a change in the buffer system of the blood had taken place. Although we have no direct information about the cause of the shift, two obvious possibilities present themselves: it could be achieved either by the addition of an acid (e.g., lactic acid) or by removal of base (e.g., by renal excretion).

Curve *III* (Fig. 2) includes data from eight of the pigeons just after panting had ceased, following their return to lower temperature. Note that the shift to the left had continued during the additional 2 hr panting between curves *II* and *III*. The  $P_{CO_2}$  had increased to 21 mm, which resulted in a return of the blood pH to near-

TABLE 1  
RESPIRATORY VOLUMES OF PIGEONS AT VARIOUS AIR TEMPERATURES

(a) Air temp. (°C)	31.5	36.6	40.3	45.0	50.9
(b) Body temp. (°C)	39.7	40.9	41.4	42.6	43.1
(c) Oxygen cons. (ml/min)	5.25	5.61	6.25	7.89	9.89*
(d) Water evap. (mg/min)	10.16	19.09	42.02	78.39	135†
(e) Minimum resp. vol (ml/min)	203	365	777	1360	2280
(f) Respiratory rate (cycles/min)	29	29	29	650	650
(g) Minimum tidal vol (ml)	7.0	12.6	26.8	2.1	3.5
(h) Arterial P <sub>CO<sub>2</sub></sub> (mm Hg)	28.5	28.5	28.5	17‡	12.3
(i) Estimated pulm. vent. vol (ml/min)	112	120	133	281	488
(k) Pulm. ventilation, % of resp. volume	55	33	17	21	21

\* Based on extrapolation of data from lower temperatures.

† Weight loss corrected for droppings and carbon dioxide loss.

‡ Interpolated from mean values for 40.0 and 50.9°C air temperature.

normal values (point *D*). If the P<sub>CO<sub>2</sub></sub> had increased to the normal 28.5 mm, the blood would have shown a low pH of 7.41 (point *E*).

Scharnke<sup>10</sup> sampled air from the interclavicular, anterior thoracic, and abdominal air sacs of pigeons. His analyses showed that the CO<sub>2</sub> content of these air sacs was 3.6–4.8 per cent (mean 4.0%) prior to exposure to high temperature, and decreased to 1.4 to 3.1 per cent (mean 2.4%) after panting. Our results make it clear that the decrease in CO<sub>2</sub> is not restricted to the nonrespiratory surfaces of the system.

*Implications for Respiratory Volumes.*—The evaporation rates observed in these birds permit the calculation of a minimum volume of air which is necessary to carry this amount of water vapor. If we assume that all evaporation takes place from the respiratory tract, and that the expired air is saturated with water vapor at body temperature, we obtain a minimum respiratory minute volume of 203 ml/min at 31°C and 1360 ml/min at 45°C [Table 1, line (e)]. The figure at 31°C compares favorably with the estimate of Salt and Zeuthen<sup>2</sup> of 150–280 ml/min. If the expired air is incompletely saturated or is at a lower temperature than the body (due to cooling at the surfaces of evaporation), the estimated respiratory volumes would be higher. On the other hand, if part of the evaporation takes place from the skin, the actual respiratory volumes would be smaller than estimated.

However, since the total evaporation increased some sevenfold between 31 and 45°C, it is most probable that the cutaneous evaporation was a negligible fraction of the total at high temperature.

A maximum respiratory minute volume (ventilation volume) of 635 ml/min in panting pigeons was measured by von Saalfeld.<sup>11</sup> This figure has been the only available estimate of minute volume in pigeons at high temperature, but the rates of evaporation which we observed suggest that the estimate was too low by a factor of at least 2. von Saalfeld's data were obtained on restrained birds with a tracheal cannula and valves, which may have reduced gas flow. Our evaporation figures were, on the other hand, obtained in unrestrained intact birds.

Tidal volume can be calculated from the estimated respiratory minute volumes and the observed respiratory rates. The respiratory rate did not change significantly as air temperature was raised from 30 to 40°C, but the evaporation and therefore the estimated tidal volume increased from 7.0 to 26.8 ml [Table 1, line (g)].

It is interesting that the maximum estimated tidal volume at 40°C air tempera-

ture, 26.8 ml, is close to the estimated total volume of the air sac system (29.4 ml according to Scharnke<sup>10</sup> and 31–60 ml according to Victorow<sup>12</sup>). When the respiratory rate at 45°C increased to a mean of 650/min, the estimated tidal volume was down to 2.1 ml. With a further increase to 50.9°C, the estimated tidal volume was 3.5 ml, still well below the tidal volume at 31.5°C. With the reservation that our calculations have a substantial margin of uncertainty, it still suggests that up to 40°C air temperature the evaporation is increased by increasing the tidal volume until the maximum obtainable tidal volume is reached. From this point on, a further increase in respiratory evaporation can take place by changing the rate. For reasons that seem connected with the economy of muscular work necessary for breathing, there is a sudden change to a stable high rate of more than 20 times the low rate. This would be analogous to the panting of dogs which, according to Crawford,<sup>13</sup> takes place at the resonant frequency of the entire respiratory system. The amount of muscular work (and heat production) necessary for respiratory movements is therefore reduced to the value necessary to keep an elastic system oscillating at its natural frequency. Since all panting takes place at a constant high frequency, a gradation of rates of evaporation can be achieved by intermittent panting and by modulation of the tidal volume. The same should also hold in other birds where panting and gular fluttering seem to occur at a constant frequency, e.g., nighthawk,<sup>14</sup> poorwill,<sup>15</sup> and ostrich.<sup>16</sup>

Since we have proposed that respiration takes place at only two distinct frequencies, and that further modulation must be in tidal volumes, it is profitable to examine how much this influences the ventilation of the lung itself and to what extent the lung can be bypassed. From the  $P_{CO_2}$  of the blood it is possible to make an estimate of the ventilation of the lung, as opposed to the ventilation of the total respiratory tract. If the blood  $P_{CO_2}$  is in equilibrium with the mean pulmonary  $P_{CO_2}$ , we can estimate the air volume into which the metabolically formed  $CO_2$  (estimated from  $O_2$  consumption and  $RQ = 0.8$ ) must be distributed to give this particular pulmonary  $P_{CO_2}$ . The estimated pulmonary ventilation at various temperatures is given in Table 1, line (*i*). Between 30 and 40°C the ventilation of the lung appears rather unchanged, i.e., in this range the increase in total ventilation [line (*e*)] seems to bypass the lung and depend only on an increased use of the air sacs. At 45 and 50°C, however, it seems that the increase in respiratory rate affects pulmonary ventilation as well. Accordingly,  $CO_2$  is lost from the blood, and alkalosis ensues.

The fraction of the total air flow which passes over the pulmonary gas exchange surfaces can be estimated from the minimum respiratory volume [line (*e*)] and the estimated pulmonary ventilation [line (*i*)]. This fraction [line (*k*)] appears to decrease as the respiratory volume increases with temperature. The decrease suggests a regulation which is compatible with the mechanism of contraction of parabronchial smooth muscles suggested by Zeuthen.<sup>3</sup>

*Conclusions.*—Pigeons can tolerate exposure to air temperatures above 50°C while maintaining body temperature at 43°C. This is achieved by evaporation of water in an amount which must correspond to the sum of metabolic heat production and heat gained from the environment. At temperatures up to 40°C, the increase in evaporation seems to be due to a deeper respiration (increased tidal volume), while respiratory frequency remains unchanged at about 30/min. There is no

concurrent change in the  $P_{CO_2}$  of the blood, which suggests that the increase in respiratory volume is achieved by the air sacs alone, without increase in the passage of air over the gas exchange surfaces of the lung. At temperatures above 45°C the respiratory frequency is increased to about 650 cycles/min, and despite a reduction in estimated tidal volume, there is loss of  $CO_2$  and considerable alkalosis. The decrease in  $P_{CO_2}$  of the blood indicates a threefold increase of the passage of air over the gas exchange surfaces of the lung.

\* Supported by NIH grants 2 TI HE 5219 and HE-02228.

† NSF graduate fellowship.

‡ NIH Research Career Award 1-K6-GM-21,522.

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**LACTATE AND PYRUVATE CONCENTRATIONS IN  
EXERCISED ISCHEMIC CANINE MUSCLE:  
RELATIONSHIP OF TISSUE SUBSTRATE LEVEL TO  
LACTATE DEHYDROGENASE ISOZYME PATTERN**

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*Communicated by Seymour S. Kety, February 1, 1966*

Lactate dehydrogenase (LDH) 1 is the most abundant of the five commonly encountered isozymes in heart, kidney, brain, and erythrocytes of many vertebrates, whereas LDH 5 is the most prominent in liver, skeletal muscle, and leukocytes.<sup>1</sup> A popular theory developed to account for this distribution maintains that LDH 5 is the principal isozyme in anaerobically metabolizing tissues because it functions more efficiently than LDH 1 at high substrate concentrations.<sup>2, 3</sup> The basis for this theory is an investigation of purified isozymes performed at 25°C with pyruvate and lactate concentrations much higher than any levels thus far shown to