Dilatation of Small Arteries and Veins in the Bat during Halothane Anesthesia

David E. Longnecker, M.D.,* and Patrick D. Harris, Ph.D.†

The microvascular responses to two concentrations of halothane in small arteries (35–45 μ) and small veins (70–100 μ) of the bat wing were quantitated in bats breathing oxygen. In addition, the anesthetic requirement of bats breathing halothane-oxygen was determined. The estimated minimum alveolar concentration (MAC) of halothane for bats was 1.14 vol per cent. The lower halothane concentration (0.81 vol per cent) was associated with significant arterial hypotension (79 ± 6 per cent of control) and small-artery dilatation (128 ± 6 per cent), but small-vein diameter was unchanged. The higher halothane concentration (1.42 vol per cent) resulted in significant arterial hypotension (53 ± 9 per cent), small-artery dilatation (148 ± 12 per cent), and small-vein dilatation (122 ± 7 per cent). These results suggest that the threshold for dilatation of small veins differs from that for dilatation of small arteries during halothane anesthesia. (Key words: Microcirculation; Small artery; Small vein; Blood pressure; Halothane; Oxygen; Anesthetic requirement; Bat.)

There are numerous reports describing the general cardiovascular effects of halothane in man1 and laboratory animals.2-4 However, information regarding the microcirculatory effects of this drug is limited. Baez and Orkin5 reported that light halothane anesthesia did not alter the dimensions of small arteries in the rat mesoappendix, while deep halothane anesthesia was associated with dilatation of both small arteries and veins. Their observations were qualitative, since no direct measurements of diameter were obtained. In addition, their animal preparation required previous basal anesthesia with pentobarbital, a drug which is associated with considerable microvascular alterations.6

The present study provides direct quantitative measurements of dimensions of small arteries and small veins in one tissue (subcutaneous) at a constant inspired halothane concentration. The common brown bat was used, because the abundant subcutaneous tissue in the wing membrane lends itself readily to light microscopy, and because it is possible to obtain quantitative data in this animal without basal anesthesia or surgical trauma.

Methods

Seventeen bats (Myotis species, random sex) provided 37 separate observations to determine anesthetic requirement in the bat. The bats were restrained gently on a glass plate which was placed in a chamber containing ports of entry and exit for the inspired gas. A mixture of oxygen and halothane was delivered to the chamber at a flow rate of 3 l/min, and gas samples from the chamber were analyzed at 5-minute intervals by gas chromatography.† After a stable concentration had been maintained for 30 minutes, the top of the chamber was removed and the foot of the bat was clamped for 30 seconds by closing a hemostat to engage the first ratchet. The presence or absence of a skeletal muscle response to this stimulus was noted and the animal was al-

*Assistant Professor of Anesthesiology and Physiology.
†Associate Professor of Physiology; Associate Investigator of the Space Sciences Research Center; Special Research Fellow 1FOHE20422 of the NIH.

Received from the Microcirculatory Systems Research Group, Departments of Anesthesiology and Physiology, School of Medicine, and the Space Sciences Research Center, University of Missouri, Columbia, Missouri 65201. Accepted for publication March 15, 1972. Supported by research grants HE12614 and 13207 from the NIH, and funds FR5387-09 and URC-702 from the University of Missouri. Portions of this material were presented at the Annual Meeting of the American Society of Anesthesiologists in Atlanta, Georgia, October 1971.

†A HCL 1673-A gas chromatograph with a single hydrogen-flame detector and a six-foot column containing 20 per cent SE-30 on 80/100 Chromosorb W at a column oven temperature of 140 C was used.
allowed to recover. Each bat was used approximately 6 to 10 days later to repeat the experiment at a different anesthetic concentration. Thus, 37 observations of "response" or "no response" were obtained.

Data from these inspired concentrations were analyzed by the technique of Behrens to estimate the minimum alveolar concentration (MAC) of halothane for bats. This technique is similar to the method used by Eger to obtain MAC in laboratory animals. Behrens' method of data analysis has been used previously to determine ethanol toxicity in bats.

Results of these observations provided a basis for selecting two anesthetic concentrations, one below MAC and one above MAC, in order to study the peripheral circulatory effects of halothane.

In the experiments which involved microcirculatory observations, nine bats (Myotis species; five females, four males; weights 5.3 to 8.7 g) were restrained gently on a glass plate which was mounted on the stage of a compound microscope as described by Nicoll and Webb. Using local anesthesia with 1/4 per cent lidocaine (total lidocaine dose less than 25 μg), the radial artery in one wing was exposed and cannulated with a small glass cannula (200–250 μ) for measurement of mean arterial blood pressure at one-minute intervals. Closed-circuit television was used to display the microscopic image of the small artery (35–45 μ) and its companion small vein (70–100 μ) on a 23-inch television monitor. The optical system contained a 12.5× objective, a 1.25× intermediate, and a 10× eyepiece lens which projected a 150× magnification of the image onto the front surface of the television vidicon. Total magnification through the optical and television systems was approximately 1,000×; thus, 1 μ in the wing appeared as a 1-mm image on the monitor. The vessel diameters were determined at 30-second intervals by marking the positions of the vessel walls on the television screen and measuring the distance between walls with a millimeter rule.

Air, 100 per cent oxygen, or oxygen plus halothane was delivered through a small-animal anesthetic system and vaporizer of our design into a lucite chamber which was placed over the bat's head and upper thorax. All inspired mixtures were delivered into the chamber at a flow rate of 200 ml/min, and excess gas escaped from the chamber through openings around the wings. A sampling port adjacent to the bat's head was used to obtain gas samples for determination of halothane concentrations by gas chromatography.

The experimental design consisted of two groups of bats. One group of four animals received an inspired halothane concentration of approximately 1.4 vol per cent, while the second group of five bats inhaled a mixture which contained approximately 0.8 vol per cent halothane. The small tidal volumes and blood volumes of these animals precluded the use of alveolar or arterial samples for halothane determination. The experimental protocol consisted of: a) a 20-minute period of inhalation of air; followed by b) a 20-minute period of inhalation of 100 per cent oxygen; c) a 45-minute period for equilibration of the anesthetic system, induction of anesthesia, and establishment of a stable inspired anesthetic concentration; and d) a subsequent 60-minute observation period, during which the animal breathed oxygen plus halothane. Induction of anesthesia was accomplished by delivering relatively high concentrations (approximately 2–2.5 vol per cent) of halothane into the breathing chamber. After the bat appeared to be anesthetized (regular respirations, absence of spontaneous motor activity, and loss of muscular tone in the extremities), the inspired concentration was reduced to 0.8 or 1.4 vol per cent halothane and the animal's condition was allowed to stabilize at either concentration for at least 15 minutes before data collection was resumed.

Data for all nine bats were pooled for the analysis of the responses to 100 per cent oxy-

§ Standard E cylinders with variable two-stage pressure regulators (Air Products, 02-1000) delivered air and oxygen through Gilmore microvascular valves into Gilmore shielded compact flowmeters. Vaporization was achieved by bubbling oxygen through the liquid anesthetic, which was contained in a 123-ml gas-washing bottle. System components were connected by 1/4-inch plastic tubing, and gases were mixed in a 4 × 5-cm airtight plastic box containing three baffles.
Figure 1 depicts the results of the 37 observations used to estimate MAC in 17 bats at room temperature. Using Behrens' method, the 50 per cent-response line (the estimated MAC) corresponds to an inspired concentration of 1.14 vol per cent halothane. Thus, the two anesthetic concentrations utilized for the microcirculatory studies were slightly above and below MAC. In the "high"-concentration group (N = 4), the inspired halothane concentration was 1.42 ± 0.03 vol per cent (mean ± SEM). The inspired halothane concentration in the "low"-concentration group (N = 5) was 0.81 ± 0.01 vol per cent. The time needed to induce anesthesia and establish a stable anesthetic concentration averaged 45 ± 4 minutes for the nine experiments.

The inhalation of 100 per cent oxygen for 20 minutes did not significantly alter small-artery diameter, small-vein diameter, or mean arterial pressure. There was a tendency for mean arterial pressure to increase to a maxi-
maximum of 108 ± 4 per cent of control, but this change was not significant. Thus, the results for observations involving halothane and oxygen were obtained by comparisons with the average values during the 20-minute period of oxygen inhalation.

Mean arterial blood pressure was reduced significantly (P < 0.05) by both concentrations of halothane (fig. 2). The mean arterial pressures 15, 30, and 45 minutes after the establishment of a stable halothane concentration were 57 ± 8, 57 ± 9, and 61 ± 10 per cent of control values, respectively, for the high-halothane group. Corresponding values for the low-halothane group were 79 ± 6, 84 ± 6, and 77 ± 3 per cent.

Changes in small-artery diameter during halothane administration are illustrated in figure 3. Both halothane concentrations resulted in significant (P < 0.05) increases in small-artery diameter. Small-artery diameters in the high-halothane group were 148 ± 13, 148 ± 13, and 148 ± 12 per cent of control at 15, 30, and 45 minutes, respectively. Corresponding values for the low halothane concentration were 129 ± 6, 128 ± 6, and 126 ± 5 per cent.

Changes in the small-vein diameters for both concentrations of halothane are depicted in figure 4. The high halothane concentration was associated with significant (P < 0.05) dilatation of the small veins; however, the low halothane concentration resulted in only statistically-insignificant diameter changes. Small-vein diameters at 15, 30, and 45 minutes were 124 ± 7, 121 ± 7, and 124 ± 8 per cent of control values, respectively, for the high concentration, and 106 ± 4, 112 ± 5, and 108 ± 3 per cent, respectively, for the low concentration.

Discussion

In our studies, it was not possible to obtain samples of alveolar gas for determination of the minimal alveolar concentration (MAC) of halothane for the bat. However, Eger, Bahlman, and Munson 13 provided a theoretical basis for estimating the alveolar concentration of an anesthetic from measurements of the inspired concentration of the drug. Their analyses suggest that within 45 minutes the alveolar concentration of halothane is at least 90 per cent of the inspired concentration in small animals. Therefore, we utilized the inspired
Fig. 3. Subcutaneous small-artery responses, as per cent of the oxygen control, in nine bats exposed to one of two concentrations of halothane in oxygen. Significant small-artery dilatation occurred at both concentrations.

Fig. 4. Subcutaneous small-vein responses, as per cent of the oxygen control, in nine bats exposed to one of two concentrations of halothane in oxygen. The higher concentration significantly dilated small veins, while the lower concentration did not alter small-vein diameter significantly.

concentration of halothane as a close approximation of the alveolar concentration in the bat.

The MAC of 1.14 vol per cent halothane in the bat correlates well with values obtained in other animals. Shim and Andersen found that the minimum alveolar anesthetic requirement of toads breathing oxygen was 0.67 vol per cent. Corresponding MAC values for the dog and man are 0.87 and 0.76 vol per cent, respectively.

In our experiments, the inhalation of 100 per cent oxygen in comparison with air did not significantly alter the observed variables. Although there is evidence that large blood
vessels do respond to changes in oxygen concentration, to date there has been no direct evidence that the inhalation of increased oxygen concentrations alters the diameters of the microvessels. Thus, it is not likely that the presence of oxygen altered the microvascular responses to halothane in our experiments.

The observed reduction in arterial pressure following the administration of halothane to bats correspond to observations in other species at similar anesthetic concentrations. Mean arterial pressures averaged approximately 57 per cent of control in the high-halothane group, while Eger et al. found an arterial pressure of 65 per cent of control in man with a similar alveolar halothane concentration at a similar time after induction of anesthesia. inspired halothane concentrations which were approximately equivalent to the high concentration used in our study resulted in reductions of arterial pressure to 58 per cent of control in miniature swine and to approximately 50 per cent of control in cats. The low halothane concentration resulted in a mean blood pressure which averaged approximately 82 per cent of control in our animals. At a similar time, Eger et al. found a mean arterial pressure of 72 per cent of control in man with a slightly higher halothane concentration.

Small-artery diameter was increased in the subcutaneous vessels of the wing membrane at both halothane concentrations. Indirect studies by other workers would indicate that these results are expected. Both Akester and Brody and Dietzel et al. have observed decreases in the vascular resistance of the skin of the dog during halothane anesthesia. Westermark has also reported decreased vascular resistance in the skin of the cat inhaling halothane. Since the arterial segment of the vascular tree is the major component of vascular resistance, the present finding of small-artery dilatation in the subcutaneous tissue of the bat wing indicates that this is a typical response to halothane in several species. Generalizations regarding small-artery diameters in other organ systems are not possible, however, since the vascular responses to halothane in other tissues may vary. Akester and Brody reported an increase in the vascular resistance of skeletal muscle during halothane anesthesia, and Baez observed no qualitative change in arterial diameters of the rat mesoappendix during light halothane anesthesia. However, both of these investigations employed basal anesthesia with pentobarbital prior to the vascular studies during halothane administration.

Small-vein dilatation was observed in this study in the high-halothane group, but the low concentration did not significantly alter the caliber of the subcutaneous small veins. Since there were significant degrees of small-artery dilatation in both groups, it appears that the sensitivity of the small veins to halothane differs from that of the small arteries. The venous response to halothane has not been well documented for other species. Clinically, the superficial veins in man appear to dilate in response to halothane anesthesia; however, Caffrey et al. were unable to demonstrate statistically significant alterations in forearm venous compliance in man during halothane anesthesia. In contrast, Morrow and Pierce reported decreased systemic venous "tonus" in dogs receiving halothane anesthesia, and Baez observed venodilatation in the rat mesoappendix during deeper levels of halothane anesthesia. However, it is difficult to compare our direct quantitative measurements of vessels in the subcutaneous tissue of the bat with qualitative observations in different organ systems and different species.

Our data do not differentiate between the possible mechanisms for the observed peripheral vascular changes. Possibilities include a direct effect on vascular smooth muscle, a neurally-mediated effect, or, more likely, a humoral mechanism related to the action of circulating vasopressin on the cutaneous vasculature, as reported recently by Akester and Brody.

In summary, we observed that inhalation of 100 per cent oxygen did not significantly alter the caliber of small arteries and veins in the bat wing. In addition, two concentrations of halothane were delivered to bats breathing oxygen. Both the low (0.81 vol per cent) and the high (1.42 vol per cent) halothane concentrations produced significant arterial hypotension and small-artery dilatation in the subcutaneous microvasculature. The subcutaneous small veins were unaffected by the low
concentration, while the high concentration resulted in small-vein dilatation. Thus, it appears that the vascular smooth muscle thresholds for dilatation are different for small arteries and small veins.

The technical assistance of Lowry Pei, Cary Hampshire, John Doyle, and David Anderson is gratefully acknowledged. The halothane used in this study was generously provided by Walter Welch, Jr., and the Ayerst Laboratories.

References