SUMMARY  The effects of halothane on intracellular membrane potential ($E_m$) and force development in cat MCA were studied. Halothane (0.07–0.14 mM/l) relaxed isolated MCA which had developed myogenic tone. Measurement of $E_m$ showed that halothane depolarized this preparation in a dose-dependent fashion in the face of vessel relaxation, demonstrating uncoupling of electrical and mechanical activity. Halothane markedly inhibited the contractile effects of histamine and serotonin suggesting that, apart from its direct action on cerebral arterial tone, it also blunts the action of vasoactive agents. When this preparation is partially depolarized from —62 to —50 mV with excess K+, halothane, while having only a small (1.2 mV) additional depolarizing effect, consistently elicits contraction rather than relaxation. Thus, the action of this particular volatile anesthetic on cerebral arteries can depend upon the resting level of $E_m$. These studies indicate that halothane relaxes myogenic tone in cat MCA by an intracellular mechanism, but that the direction of its effect (i.e., relaxation vs. contraction) may depend upon the prior level of $E_m$ and muscle cell activation.

With respect to the cardiovascular system, the predominant effects of halothane anesthesia are depressor. Halothane inhibits myocardial functions and decreases peripheral vascular resistance. The direct depressor actions of halothane on cardiac muscle appear to involve alterations in intracellular Ca$^{2+}$ handling and reduction of Ca$^{2+}$ availability to contractile elements; such actions may involve depression of sarcoplasmic reticulum (SR) function. Similarly, in sinoatrial (SA) nodal tissue, halothane exerts negative chronotropic actions which can be partially attributed to reduction of inward current during the action potential and partially due to inhibition of intracellular Ca$^{2+}$ availability.

Halothane increases cerebral blood flow and can increase intracranial pressure demonstrating its potent cerebral vasodilatory action. However, it is not known whether such effects are direct or result from changes in cerebral metabolism and/or inhibition of normally occurring vasoactive neurotransmitters or hormones. Halothane can also abolish cerebrovascular autoregulatory responses to changing arterial blood pressure.

Halothane, like most volatile anesthetics, increases membrane fluidity in a variety of cell types. This action of halothane suggests that it can exert direct effects on muscle cells by a mechanism involving changes in membrane potential and in ionic permeabilities. It is the purpose of these studies to determine the direct action of halothane on mechanical and electrical properties of isolated cat middle cerebral artery (MCA) in order to more clearly define its potent dilatory action on cerebral arteries, and to determine if the predominant actions occur through membrane electrical events or via intracellular mechanisms.

Methods

Adult mongrel cats (2.5–4.0 kg) of either sex were anesthetized with sodium pentobarbital (30 mg/kg) and decapitated. The brain was removed and placed in cold, oxygenated Kreb’s solution. MCA were removed and used for study. Three mm segments of artery were mounted in a specially fabricated myograph to record force development. The lumen of the segment was threaded with 2 pieces of 22 um tungsten wire. The wires were stretched over the open jaws of 2 stainless steel rings. One ring was anchored and the other connected to a very sensitive load cell (Kulite Semiconductor Products, Inc.; Ridgefield, N.J.). Under control conditions the preparation was continually suffused with a physiological salt solution containing (in mM): Na$^+$ 141, K$^+$ 4.7, Cl$^-$ 125, Ca$^{2+}$ 2.5, Mg$^{2+}$ 0.76, H$_2$PO$_4$ 1.7, HCO$_3^-$ 25, glucose 11 and HEPES (N-2-hydroxy-ethylpiperazine-N-2-ethane-sulfonic acid) 5. Solutions were aerated with 95% O$_2$/5% CO$_2$, giving a pH of 7.35–7.42 and POCO$_2$ of 35–40 torr. Temperature was maintained at 37°C ± 0.2°C. Halothane (2-bromo-2-chloro-1,1,1-trifluoroethane without 0.1% thymol) was aerated into the solution via a Draeger Vaporizer at 1.5 to 2.5 vol. %. This level of halothane vaporization gave levels of 0.07 to 0.14 mM/l analyzed on a gas chromatograph. Arterial segments were placed under 400 mg passive load and allowed to equilibrate for 90 min prior to study, during which time they developed between 200 and 400 mg of myogenic tone. Histamine ($10^{-7}$ to $10^{-3}$ M) was added to the bathing solutions in cumulative doses.

For measurements of $E_m$, glass microelectrodes were used according to techniques previously described. They were filled with 3 M KCl and had tip resistances between 50 and 80 meqohms, with tip po-
CEREBRAL ARTERIAL EFFECTS OF HALOTHANE

Inhibitory Action of Halothane on Middle Cerebral Artery

Middle cerebral arterial segments were mounted in a muscle myograph and equilibrated with a passive load. Ninety percent of the preparations developed myogenic tone ranging from 100 to 400 mg. As can be seen in figure 1, halothane at 2.0 and 2.5 vol. % relaxed these arterial segments. Similar responses were observed in all ten (100%) vessels studied in this manner.

To determine the effects halothane might have on drug induced responses of cat MCA, dose-response curves for histamine-induced mechanical activity were done in the presence and absence of halothane (1.5 vol. % or 0.07 mM/L). A typical experiment in this regard is depicted in figure 2. As can be seen, halothane markedly inhibits both the relaxant (occurring at 10⁻⁶ and 3 × 10⁻⁶ M) and contractile (10⁻⁵ to 10⁻³ M) actions of histamine. Quantitation of six such experiments (representing 94% of all preparations studied in this manner) is given in figure 3. Halothane abolishes the relaxant actions of histamine and decreases both sensitivity and contractility to histamine effects.

Effect of Halothane on the Membrane Potential (E_m) of Cat Middle Cerebral Artery

E_m was measured intracellularly with glass microelectrodes from muscle cells of cat MCA. The control E_m was -62 ± 2.0 mV. Halothane at 1.5 and 2.0 vol. % (0.07 to 0.14 mM/L) significantly depolarized all of the 7 preparations studied to a mean value of -53 ± 2.1 and -45 ± 1.6 mV respectively (fig 5). This finding demonstrates an uncoupling of electrical and mechanical events by halothane in this preparation in that the vessel relaxation is occurring while the muscle cells are depolarizing. Taken together, such data suggest that halothane, while having direct effects on ion permeabilities resulting in reduction of E_m, induces relaxation via an intracellular mechanism. If halothane worked via electromechanical coupling in this vessel the membrane depolarization would result in muscle activation, not the observed relaxation.

However, when MCA are partially depolarized and activated by 30 mM K⁺ (corresponds to an E_m of -46 ± 2.0 mV and 25% maximum activation) halothane...
The results of this study demonstrate that halothane inhibits myogenic tone and significantly reduces the contractile effects of histamine and serotonin. The effect of halothane on myogenic tone appears to work via an intracellular mechanism rather than through electromechanical coupling. Uncoupling of electrical and mechanical activity (i.e., depolarization in the face of contraction) is a unique property of halothane in that we are unaware of similar actions mediated by any other pharmacological agents. It would appear that the action of halothane in increasing cerebral blood flow may work through direct inhibitory actions on arterial muscle. The data presented here do not allow direct comparison of the intracellular effects of halothane on Ca\(^{2+}\) availability in cardiac tissue with those observed in cerebral arteries, but may prompt future experiments in this regard.

Halothane can destroy autoregulatory behavior in the face of changing arterial blood pressure. We have found that myogenic autoregulation of cerebral arteries is dependent upon changes in $E_m$ of smooth muscle cells (i.e., electromechanical coupling). The finding that halothane uncouples electrical and mechanical events may provide a cellular mechanism through which attenuation of cerebral blood flow autoregulation by halothane occurs.

Our finding that halothane contracts cerebral arteries which are partially depolarized is difficult to reconcile with its inhibitory action on myogenic tone. Depolarization of arterial muscle usually results in activation by increasing Ca\(^{2+}\) conductance through voltage dependent channels. It may be that once Ca\(^{2+}\) channels are activated by partial depolarization with excess K\(^+\), halothane can then augment conductance through these sensitive ionic channels. If such a hypothesis is indeed correct, it may be linked to halothane's action of changing membrane fluidity, there-
by, altering ionic permeabilities.\textsuperscript{11,12} An increase in membrane fluidity by halothane may result in the observed depolarization through inhibition of ionic conductances, such as K\textsuperscript{+}, by not allowing the channel to remain in a fixed configuration within the membrane. Obviously, more experiments are needed to clarify the direct action of halothane on the plasma membrane.

Since the action of halothane on cerebral arterial muscle appears to be modified by the level of E\textsubscript{m}, its effect on cerebral blood flow may depend upon metabolic and/or humoral conditions which might regulate resting E\textsubscript{m} in this tissue (e.g., cerebral spinal fluid K\textsuperscript{+}, adenosine or neural activity). Such a mechanism may help explain the wide variability in cardiovascular responses to halothane anesthesia in humans.

References

Cellular actions of halothane on cat cerebral arterial muscle.
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