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Volatile Anesthetic Agents and Vascular Communication in the Microcirculation of the Hamster Cheek Pouch

Patricia Satitpunwaycha, M.D., Richard J. Rivers, M.D., Ph.D.†*

Background: **There is communication between tissue and the vascular network involved in regulating distribution of blood flow. Signals generated by the tissue are communicated upstream to create a coordinated network response in unison with other controllers of blood flow, such as myogenic and flow-dependent responses.**

Methods: **This vascular communication was modeled with the microapplication of methacholine (10^{-4} M) or potassium chloride solution (KCl; 100 mM) to arterioles (40–60 μm in diameter) of the cheek pouch of anesthetized hamsters and viewed with videomicroscopy. Local and conducted (500 μm upstream) responses were measured. Halothane or isoflurane (1%, 2%, and 3%) was equilibrated with the superfusion solution and applied to the entire tissue. Responses to KCl and methacholine were then repeated in the presence of an anesthetic agent.**

Results: **Halothane and isoflurane increased the resting diameter of the arterioles. They also decreased the methacholine-initiated dilations. To test for the effects of increased resting diameter on the dilations, 0%, 5% and 10% oxygen alone was applied to the pouch to alter the tone, and the methacholine responses were repeated. The dilations decreased with oxygen-induced increases in resting diameter, but the conducted dilation decreased to a lesser extent than was seen with the volatile anesthetic agents. Neither halothane nor isoflurane decreased constrictions caused by KCl.**

Conclusions: **Decreased methacholine-initiated conducted dilations caused by halothane and isoflurane were not due to decreases in cell–cell communication because KCl conducted responses persisted. Therefore, cell–cell vascular communication appears intact in the presence of clinical concentrations of halothane and isoflurane. (Key words: Arterioles; conducted vasodilation; halothane; isoflurane; muscarinic agonist.)**

CONTROL of distribution of blood flow to the tissues is a multilevel process, which contributes to the integration of vasomotor tone in the local regulation of blood flow.[1] In conduit arteries, individual factors such as pressure, flow, and neural processes interact to bring blood to the tissues.[2] In the terminal vasculature, however, a different mechanism, known as the conducted vasomotor response, appears to be significant in the coupling of local metabolic stimuli to dilation of upstream arteriolar segments,[3] thereby matching the resistances of the vascular segments with the local need for tissue perfusion. Cell-cell communication of electronic signals through gap junctions is thought to be the mode of the communication.[4]

Halothane has been reported to uncouple gap junctions and thereby potentially interfere with intercellular communication. Burt and Spray demonstrated that halothane decreased gap junctional conductance in myocytes by decreasing the number of conducting channels (rather than reducing the unitary conductance of individual channels).[5] This reversible inhibition appears to correlate well with the octanol/water partition coefficient of halothane and other halocarbons, suggesting that membrane incorporation and modification of the immediate environment around gap junctions is a possible mechanism.[6] Given the ability of halocarbons to inhibit gap junctions and the likely role for the latter in intercellular communication in the peripheral vasculature, our aim was to determine if clinical concentrations of halothane and isoflurane could affect conducted responses.

Materials and Methods

Preparation of Hamster Cheek Pouch and Video Microscopy

Male golden hamsters (100-150 g) were anesthetized with sodium pentobarbital (70 mg/kg given intraperito-

* Anesthesiology Resident, Johns Hopkins University.

† Associate Professor, Departments of Anesthesiology and of Pharmacology and Physiology, University of Rochester.

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Address reprint requests to Dr. Rivers: Department of Anesthesiology, University of Rochester Medical Center, 601 Elmwood Avenue, Rochester, New York 14642. Address electronic mail to: rrivers@anes.rochester.edu

neally; Nembutal; Abbott, North Chicago, IL) tracheotomized, and maintained at appropriate hydration and depth of anesthesia by continuous intraperitoneal injection of saline (0.008 ml/min) and pentobarbital (0.075 mg/min). The intact cheek pouch was rinsed with distilled water, exteriorized, and superfused with 37.5°C physiologic saline solution (128 mM NaCl, 4.7 mM KCl, 2.0 mM $CaCl_2$, 1.2 mM $MgSO_4$, 5 mM glucose, 2 mM pyruvate, 18 mM $NaHCO_3$, 1.2 mM NaH_2PO_4, and 0.02 mM ethylenediaminetetraacetic acid) equilibrated with 0%–10% O_2/5% CO_2 with balance N_2 at a rate of ≈5 ml/min. An intravital microscope (Wild Kombistereo microscope, Leica, Inc., Deerfield, IL) coupled to a video camera (Dage CCD72, Dage-MTI Inc., Michigan City, IN) was used to view transilluminated pouch arterioles. A manually controlled video micrometer (Microcirculation Research Institute, College Station, TX) was used to measure the inside arteriolar diameters in real time.

After dissection to clear overlying connective tissue, the preparation was allowed to stabilize for at least 60 min.[7] Vessels 40–60 μm in maximum diameter were selected for study. The maximum diameter of the arteriole was determined by applying a few drops of 10^{-2} M methacholine directly onto the tissue. Viability of the preparation was assessed by observing a vasoconstrictor response when superfusate oxygen concentration was elevated from 0% to 5%. In the presence of 5% O_2, all blood vessels studied had spontaneous resting diameters of 40–60% of the maximum diameter. Deep esophageal temperature was maintained at 37°C by a combination of conductive and radiant heating.

Application of Drug and Data Collection

Methacholine (Sigma Chemical Co., St. Louis, MO) was prepared as a 10^{-2} M stock solution in bicarbonate buffered solution and brought to the test concentration each day with additional solution. Methacholine (10^{-4} M) was pneumatically (Narishige Microinjector, model IM-200, Sea Cliff, NY) applied *via* micropipette (1–2 μm ID) for 5 s to cause dilations locally, at the tip of the pipette, and dilations conducted beyond the local application of the drug. The tip was placed outside the adventitia of the vessel, 10–15 μm from the vessel wall. Drug was ejected pneumatically with the minimum pressure that would create the maximum response (1–2 lb/in^2) as previously described.[8] Measurements of arteriolar diameter were taken at the site of drug application (local response) and a remote site 500 μm upstream (conducted response). The response was viewed sequentially at each location, using repeated applications of methacholine.

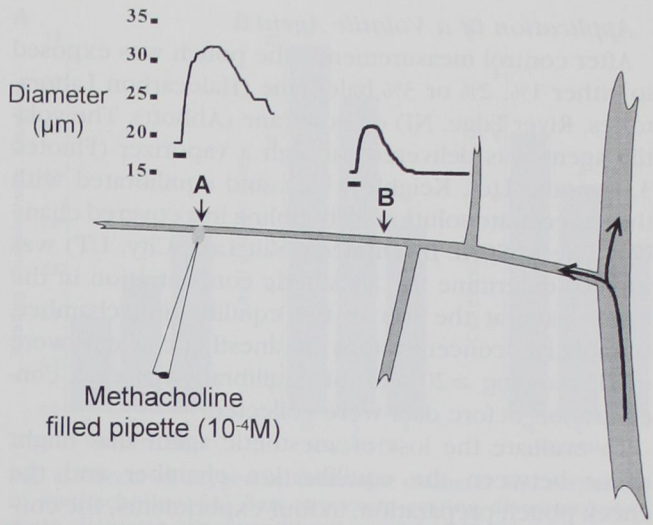

Fig. 1. Conducted dilation is modeled through the application of methacholine *via* micropipette. Drug is applied for 5 s (*solid bar*) at one location, and responses are recorded at locations along the arteriole that are remote (500 μm upstream) from the site of application. Typical dilations caused by the application of methacholine are shown.

One to three repeated measurements were made at each location and then averaged. The sample responses depicted in figure 1 are an accurate representation of the duration of the response as drug was washed away by the superfusate after the 5-s ejection. To test for convection of the drug to remote sites, the drug also was applied at a location in the tissue 500 μm from the arteriole. Conducted responses at the remote site were seen only when drug was applied directly onto the arteriole. Previous studies have ruled out contributions of diffusion using this technique.[8] Calculations of diffusion time to the remote site show that the time necessary for diffusion is an order of magnitude greater than the onset time of the response.

KCl (100 mM) was prepared by exchanging potassium for sodium in the bicarbonate buffered solution. It was applied pneumatically for 5 s through micropipettes with the minimum pressure that would create the maximum local response (10–30 lb/in^2). Pressure ejection caused obvious movement of the arteriole with all applications. In control studies with normal bicarbonate solution, however, this method of application had no effect on the arteriolar diameter. Tests for convection of KCl from the micropipettes to the remote locations also were performed as previously described, and there was no evidence that high concentrations of KCl were reaching the remote location.

Application of a Volatile Agent

After control measurements, the pouch was exposed to either 1%, 2% or 3% halothane (Halocarbon Laboratories, River Edge, NJ) or isoflurane (Abbott). The volatile agent was delivered through a vaporizer (Fluotec 3, Cyprane Ltd., Keighley, UK), and equilibrated with the superfusate solution by bubbling in a covered chamber. The RASCAL II (Ohmeda, Salt Lake City, UT) was used to determine the anesthetic concentration in the head space at the top of the equilibration chamber. Cumulative concentrations of anesthetic agent were used, allowing ≥20 min for equilibration at each concentration before data were collected.

To evaluate the loss of anesthetic agent that might occur between the equilibration chamber and the cheek pouch preparation, in four experiments, the concentration of halothane was measured by high-performance liquid chromatography. The procedure for direct determination of volatile anesthetic agents in buffer solution was adapted from the literature.[9] For preparation of the calibration graph, various amounts (4.11, 8.23, and 16.5 mg) of halothane were measured into gastight syringes and mixed with 100 ml buffer solution to obtain final concentrations of 0.21, 0.42, and 0.84 mM halothane. Five hundred microliters of the sample was added to 500 μl internal standard mixture (0.05 nM toluene in methanol), and 20 μl of the final sample was injected into the column. Samples were then taken from both the equilibration chamber, while bubbling with 2% halothane, and the solution after it had finished flowing over the cheek pouch preparation. Five hundred microliters of each sample was added to the 500 μl of internal standard and treated as described previously. Concentrations of isoflurane were not determined.

The chromatographic system consisted of G2S5 LC system (Waters, Bedford, MA) and Chromanetics C18 hypersal ODS, 4.6 cm × 100 mm, 3 μm spherical particles (ANSPEC, Ann Arbor, MI). The chromatograms were recorded and processed by Rainin Dynamax software.

In six hamsters, the systemic blood pressure was measured through a catheter in the femoral artery. Blood pressure was measured before (baseline pressure 100 ± 23 mmHg) and after halothane (3%) or isoflurane (3%) were applied to the pouch preparation for 20 min. Applying an anesthetic agent to the pouch in this fashion was found to have minimal effect on the systemic pressure (90 ± 17, 87 ± 14 mmHg).

The anesthetic concentrations, set according to the head space analysis, are normalized to minimum alveolar concentration in humans. Minimum alveolar concentration in hamsters was not determined because the hamsters were not anesthetized with these agents.

Control for Resting Diameter

Because halothane and isoflurane each increased arteriolar diameter, a series of experiments was performed to test for the direct effect of resting diameter on conducted dilation. Differing resting diameter was achieved by using different concentrations of oxygen in the superfusion solution. Cheek pouch preparations were exposed to 0%, 5%, and 10% O_2, in random order, and allowed to equilibrate for 20 min. Conducted dilations were then measured using methacholine to initiate the response, as described previously. In these control experiments, no volatile anesthetic agents were applied to the tissue preparation.

Data Analysis

Resting diameter is defined as the average diameter of the arteriole for the 4 s before each application of methacholine or KCl. The resting diameter was stable for ≥30 s before any test was performed. The response was the maximum change in diameter from baseline that was recorded during the 1-min interval after application of drug. Data are presented as absolute changes in diameter, as diameters normalized to the maximum vessel diameter, or as responses normalized to control conditions. The data were analyzed using analysis of variance with repeated measures when appropriate. If a significant difference was found, individual concentrations or conditions were compared with the control values using JMP Statistical Software (SAS Institute, Inc., Cary, NC). Data analyzed by comparing means are presented as means ± SEM. Data identifying populations are presented as means ± SD. Findings were considered significant if $P < 0.05$.

Results

Volatile Anesthetic Agents

Isoflurane (n = 6; maximum arteriolar diameter, 50 μm ± 3.9) and halothane (n = 14; maximum arteriolar diameter, 48 μm ± 5.4) were studied in separate sets of experiments using 5% O_2 in the superfusate. Each agent demonstrated dose-dependent vasodilation of arterioles across the tissue with this tissue-wide applica-

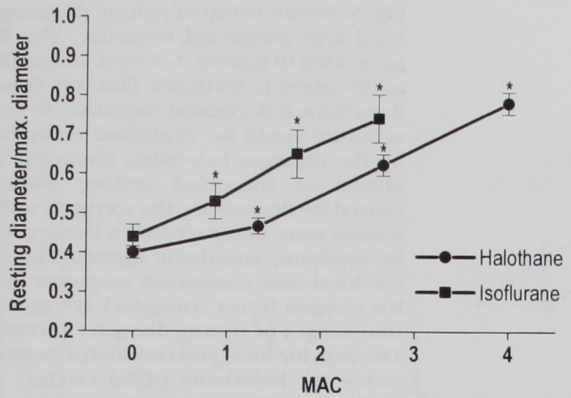

Fig. 2. Effects of anesthetic agents on arteriolar diameter. Points represent arteriolar resting diameters normalized to the maximum diameter and were measured just before application of methacholine. There were significant dose-dependent vasodilatory effects of both volatile anesthetic agents. *Significantly different from control (0 minimum alveolar concentration; isoflurane, $P < 0.0001$; halothane, $P < 0.0001$).

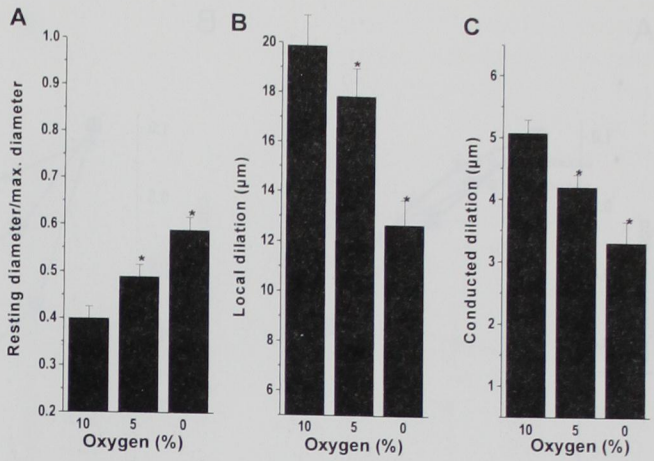

Fig. 4. Effects of oxygen on arteriolar diameter and responses to methacholine. (*A*) Bars represent resting diameter/maximum diameter after equilibration with differing concentrations of oxygen and before application of methacholine. Vessel tone significantly decreased with decreasing oxygen concentration ($P < 0.0001$). (*B*) Bars represent average local response (in micrometers) to the application of methacholine. Local responses decreased with decreasing oxygen concentrations ($P < 0.0001$). (*C*) Bars represent average conducted responses (in micrometers) at the 500-μm upstream location in response to methacholine. Conducted response decreased with decreasing oxygen concentrations ($P < 0.0001$).

tion (fig. 2). Vessels exposed to 3% anesthetic agents had such substantial increases in resting diameter that there was not always sufficient dilatory capacity to detect additional dilation when methacholine was applied. Therefore, dilation data collected while using 3% of either agent are not presented. Only conducted constrictions could be measured reliably when the anesthetic agents were at 3%. These data are presented in the subsequent sections.

Conducted Dilation

Each anesthetic agent attenuated the local and conducted dilations induced by methacholine (fig. 3). All effects were reversed completely when the volatile anesthetic was discontinued (data not shown).

Oxygen and Resting Diameter

Superfusion oxygen concentration affected arteriolar resting diameter (n = 10; maximum arteriolar diameter,

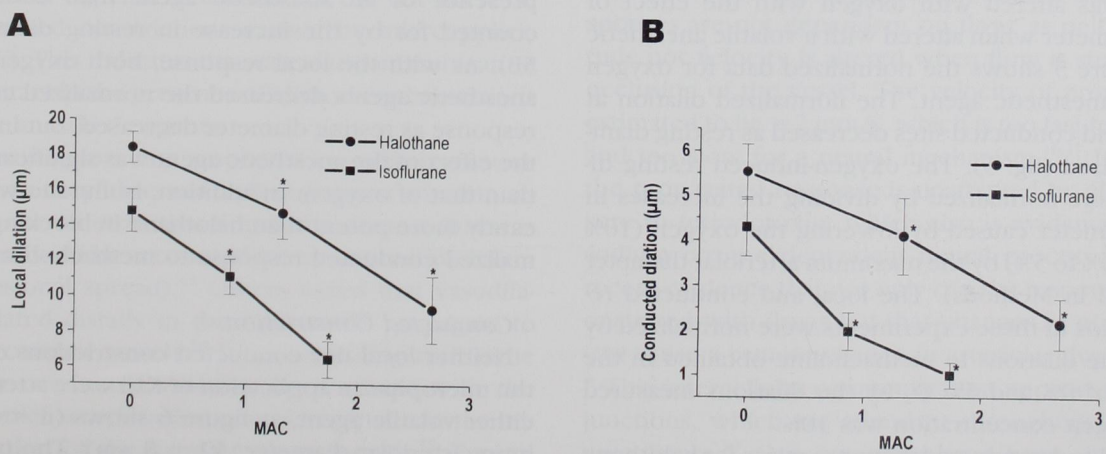

Fig. 3. Effects of anesthetic agents on arteriolar responses to methacholine. Points represent vasodilation in response to methacholine. Both local (*A*) and conducted responses 500 μm away (*B*) are significantly decreased after equilibration with volatile anesthetic agents. *Significantly different from control (isoflurane, $P < 0.0001$; halothane, $P < 0.0001$).

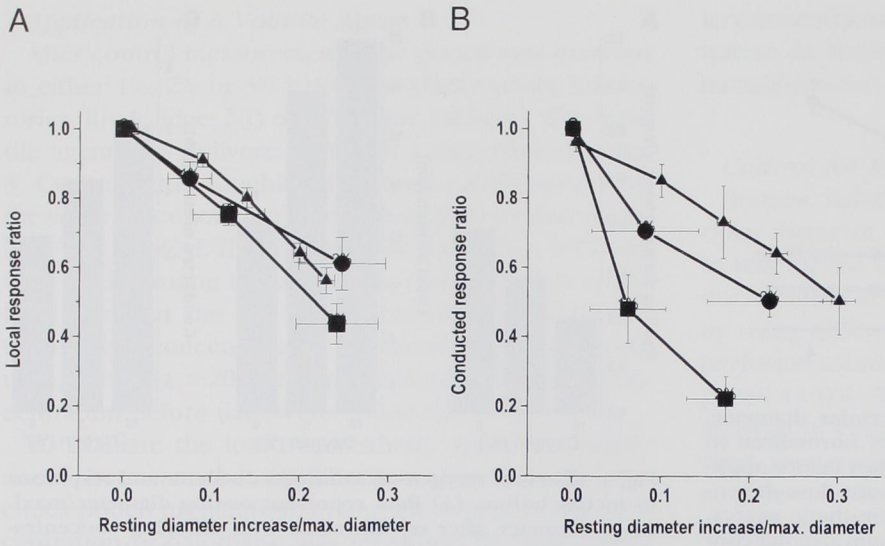

Fig. 5. Relationship of resting diameter to local and conducted response. The data presented in figures 2–4 were normalized as discussed in methods. This was done to determine if decreased responses to methacholine could be explained completely by the changes in resting diameter. The effects of increased resting diameter caused by decreasing the oxygen concentration were compared with those caused by applying anesthetic agents. The data for local and conducted response ratios for oxygen (*open triangles*) are grouped into ranges of resting diameter increases. The data for local and conducted response ratios for halothane (*filled circles*) and isoflurane (*filled squares*) are grouped by anesthetic concentration (C = control, 1%, 2%). (*A*) Normalized local dilations caused by methacholine *versus* normalized changes in resting diameter. The effect of resting diameter was not significantly different between any treatments (oxygen, halothane, or isoflurane). (*B*) Normalized conducted dilations *versus* normalized changes in resting diameter. Analysis of variance of these normalized data showed significant differences in the way the resting diameter affected the conducted response ($P = 0.0063$) depending on the treatment. Therefore, volatile anesthetic agents affected conducted dilations significantly more than could be explained by a changes in resting diameter (as defined by lower oxygen).

52 ± 7 μm). Diameter increased from ≈40% to 60% of the maximal arteriole diameter when oxygen concentration changed from 10% to 0% (fig. 4). There was also a significant decrease in both the local and the conducted vasomotor responses to methacholine when oxygen concentration was decreased.

It is has been shown previously that responses of arterioles to agonists depends on the initial value of vascular tone.[10–12] To quantify the effect of initial tone, as estimated by resting diameter, on the dilatory responses, we compared the effect of resting diameter when it was altered with oxygen with the effect of resting diameter when altered with a volatile anesthetic agent. Figure 5 shows the normalized data for oxygen and each anesthetic agent. The normalized dilation at the local and conducted sites decreased as resting diameter increased (fig. 5). The oxygen-induced resting diameters were normalized by dividing the increases in resting diameter caused by lowering the oxygen (10% to 0% or 10% to 5%) by the maximum arteriolar diameter (as defined in Methods). The local and conducted response ratios in these experiments were normalized by dividing the dilations to methacholine obtained in the presence of 0% and 5% O_2 by the dilations measured when oxygen concentration was 10%.

The local and conducted response ratios for halothane and isoflurane were normalized by dividing the methacholine-induced dilations by the dilations obtained in the absence of an anesthetic agent. The anesthetically induced changes in resting diameter were normalized by dividing the increase in resting diameter by the maximum arteriolar diameter. Overlap of lines in figure 5A signifies that there was no difference among treatments (oxygen, halothane, and isoflurane) in their effects on vessel response at the local site. The decreased methacholine-induced dilation at the local site may be explained completely by the decreased resting diameter.

In contrast to the local responses, however, the conducted responses decreased significantly more in the presence of an anesthetic agent than could be accounted for by the increase in resting diameter (fig. 5B). As with the local response, both oxygen and the anesthetic agents decreased the normalized conducted response as resting diameter decreased, but in this case the effect of the anesthetic agent was significantly more than that of oxygen. In addition, isoflurane was significantly more potent than halothane in blocking the normalized conducted response to methacholine.

Conducted Constriction

Neither local nor conducted constrictions caused by the micropipette application of KCl were attenuated by either volatile agent, as figure 6 shows (n = 11; maximum arteriolar diameter, 52 ± 8 μm). The conducted constrictions were significantly increased in the presence of either agent. The effect of increased resting

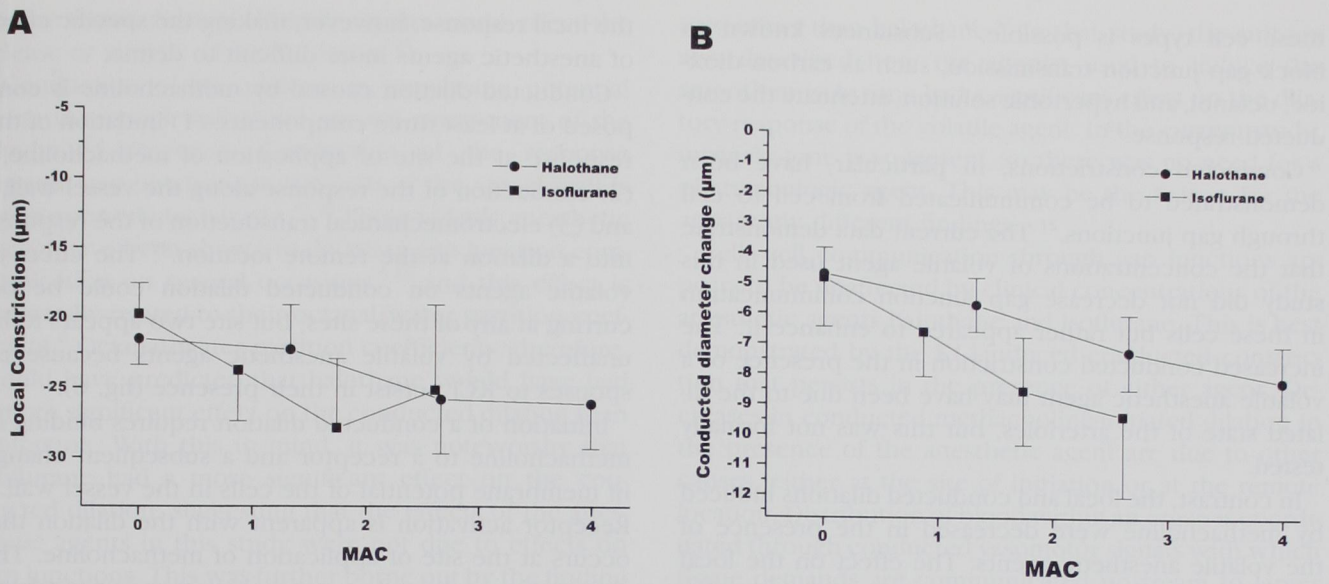

Fig. 6. Effect of anesthetic agents on local and conducted vasoconstriction caused by the microapplication of KCl. Neither halothane nor isoflurane decreased the constrictions caused by KCl. Note that the axis shows negative values because the diameter became smaller with applications of KCl.

diameter *per se* on the constriction responses was not determined because the volatile agents did not decrease the response.

Discussion

This study shows that in the microcirculation of the hamster cheek pouch, volatile anesthetic agents decreased both the local dilation and conducted dilation caused by the microapplication of methacholine in a concentration dependent fashion but did not decrease the conducted constrictions caused by microapplication of KCl. This indicates that vascular communication remains intact in the presence of clinical concentrations of volatile anesthetic agents.

Early investigators first suspected vascular communication when they observed a phenomenon wherein responses to vasomotor stimuli spread to an area that was larger than could be accounted for by direct stimulation (*i.e.,* diffusional spread).[13] Others noted that vasodilations initiated distally in the limb could propagate to proximal conduit vessels.[14] In 1970, Duling and Berne induced a conducted vasodilatory response with the application of acetylcholine *via* micropipette to the microcirculation of the hamster cheek pouch.[15] Typical experimental responses are shown in figure 1. Subsequent studies during the past 25 yr have helped to characterize this mechanism for arteriole to feed vessel (cell-cell) communication.

Conducted vasomotion has been described as a bidirectional spread of a vasomotor response from a specific site of initiation.[15] The signal appears to be conducted along the vessel wall and has properties consistent with that of an electrical current in a cable, decaying exponentially with time and distance. Likewise, the responses are additive in amplitude and direction: Dilation or constriction initiated at different locations sum when they arrive at a common branch. Conducted responses are not dependent on flow, as neither magnitude nor velocity is altered when flow is stopped after occlusion of the vessel. The velocity of propagation is estimated to be $\geq$2 mm/s, which is too fast for diffusion and too slow for a neural mechanism.[16,17] In addition, the propagated response is unaffected by global exposure to tetrodotoxin, which also is evidence against a sodium channel-dependent neural process.[18] There is recent evidence that not only can the response be demonstrated with drugs but that changes in pressure also can cause a remote change in arteriolar diameter.[19]

This intercellular communication appears to use gap junctions, which are abundant throughout the microvasculature.[20] Gap junctions have been described in both endothelial cells and vascular smooth muscle cells; therefore, electrical communication within or between

these cell types is possible.[21] Substances known to block gap junction transmission, such as carbon dioxide, octanol, and hypertonic solution, attenuate the conducted response.[22]

Conducted constrictions, in particular, have been demonstrated to be communicated from cell to cell through gap junctions.[21] The current data demonstrate that the concentrations of volatile agent used in this study did not decrease gap junction communication in these cells but rather appeared to enhance it. The increased conducted constriction in the presence of a volatile anesthetic agent may have been due to the dilated state of the arterioles, but this was not formally tested.

In contrast, the local and conducted dilations induced by methacholine were decreased in the presence of the volatile anesthetic agents. The effect on the local response could be explained completely by the simultaneous decreased resting diameter. Vasomotor responses are well known to be dependent on resting vascular tone.[10-12] In the current vessels, the loss of local response to methacholine caused by the presence of volatile agent closely matched the loss of response caused by lowering the concentration of oxygen and the resultant increase in resting diameter.

Both volatile agents have been shown previously to affect the relaxation caused by acetylcholine in rings of arteries. This inhibition was shown to be due to an effect on endothelium-dependent dilations caused by production of nitric oxide[23] or EDHF.[24] These findings appear to be closely related to the findings regarding the local response described in this study; however, the decreased local dilation seen in the presence of the anesthetic agents described in our study also could be explained completely by the changes in arteriolar resting diameter. Nitric oxide or EDHF may have been playing a role in the local response, but it was not demonstrated here.

In sharp contrast to the effects of anesthetic agents on the local dilations, however, the volatile agents had significantly more effect on the conducted dilation than could be explained by the increase in resting diameter. Compared with the decreased response noted in the presence of lower concentrations of oxygen, the volatile agents had a significantly greater effect. This supports the finding that the mechanisms of conducted dilations are unique from those of local dilations,[25,26] and therefore volatile agents could have had differential effects. The conducted response is more complex than the local response, however, making the specific effect of anesthetic agents more difficult to define.

Conducted dilation caused by methacholine is composed of at least three components: (1) initiation of the response at the site of application of methacholine,[27] (2) conduction of the response along the vessel wall,[28] and (3) electromechanical transduction of the response into a dilation at the remote location.[29] The effect of volatile agents on conducted dilation could be occurring at any of these sites, but site two appears to be unaffected by volatile anesthetic agents because responses to KCl persist in their presence (fig. 6).

Initiation of a conducted dilation requires binding of methacholine to a receptor and a subsequent change in membrane potential of the cells in the vessel wall.[28] Receptor activation is apparent with the dilation that occurs at the site of application of methacholine. The size of the conducted response, however, is not related to the local response. Conducted responses have been reported even when a local response was absent.[30] Therefore, intracellular mechanisms creating the local and conducted responses are not necessarily related. The current data confirm that multiple mechanisms of arteriole dilation are being activated through the single application of a muscarinic agonist, and it is possible that different muscarinic receptor subtypes are involved in the two responses.[31-33] Therefore, although the local response implies that muscarinic receptor activation was intact for the local response, the receptors causing the conducted response may have been affected differentially. The effect of the these volatile agents on specific muscarinic receptor subtypes or the ability to cause a change in membrane potential has not been determined. Previous reports that volatile anesthetic agents can disrupt muscarinic agonist G-protein coupling may offer a pathway whereby the conducted dilations are decreased.[34]

A change in membrane potential after muscarinic receptor activation may occur directly through receptor[35,36] or through the release of a substance from the endothelium that causes changes in cell membrane potential. Nitric oxide and endothelium-dependent hyperpolarizing factor are two candidates that could be released from the endothelium to hyperpolarize the cells. Nitric oxide is not likely, however, because brief local application of nitric oxide itself, *via* nitroprusside, does not cause a conducted response.[37] Endothelium-derived hyperpolarizing factor, in contrast,[31] may be a better candidate, but because its identity is unknown, it can-

not be tested directly. Volatile agents could affect the release or activity of such a factor.[24]

Conduction of the change in membrane potential along the vessel wall is the second component of the conducted response. Conduction of the response through gap junctions in the cells of the vessel wall is the purported mechanism.[21,28] These volatile anesthetic agents have been shown to decrease gap junction communication on several occasions,[5,38] and this effect is reportedly related to their octanal/water partition coefficient.[6] Octanal/water partition coefficients, therefore, would have predicted that halothane would have had a more significant effect on the conducted dilation than isoflurane. With this in mind, it was noteworthy that isoflurane had a more significant effect on the conducted dilation, suggesting that the effects of the anesthetic agents in this study were not due to effects on gap junctions. This was further borne out by the finding that the conducted constrictions created with KCl also were not decreased in the presence of the volatile anesthetic agents. Therefore, the mechanism whereby these volatile agents affect conducted dilation is not due to an effect on gap junction communication between the cells.

The final component of the pathway to a conducted response is electromechanical transduction of the conducted signal into a dilatory response.[29] This may be directly due to voltage-sensitive calcium channels being closed, thereby causing relaxation of the smooth muscle. Alternatively, the changes in membrane potential cause a substance to be released that in turn causes the dilation.[39] Release of nitric oxide seems unlikely, however, because the conducted component of the response persists in the presence of nitroarginine.[26]

An effect of volatile anesthetic agents on electromechanical transduction has not been formally studied, but there is evidence that halothane or isoflurane can block voltage-gated calcium channels[40,41] that are important components of the electromechanical response. This could explain the direct dilation created by volatile agents and the decreased conducted dilation.

Dilation of Arterioles by Anesthetic Agents

This study demonstrates that volatile anesthetic agents directly dilate the arteriole beds of the hamster cheek pouch. Isoflurane was a more potent dilator than halothane when compared on the arbitrary minimum alveolar concentration basis used here. This is in contrast, however, to findings on isolated porcine coronary segments, in which isoflurane had significantly less dilatory effect than halothane.[42] In that study, the authors also described how the agonist used to induce the smooth muscle tone had a significant effect on the dilatory response of the volatile agent. In the current study, intrinsic tone was present, so there was no need for a pharmacologic agent. This may be the reason for the apparently different findings.

Cell-cell communication through gap junctions appears to be unaffected by clinical concentrations of the anesthetic agents halothane and isoflurane. This is best demonstrated by the KCl-induced conducted constriction that persists in the presence of either agent. Decreases in conducted methacholine-initiated dilation in the presence of the anesthetic agent are due to other causes, either at the site of initiation or at the remote location. Distribution of blood within an organ is coordinated through conducted vasomotor signals with which tissue demands are communicated upstream to larger vessels. If this communication is altered, then regions of tissue demanding additional blood theoretically may not be able to communicate this demand to the feed vessels. In times of severe physiologic stress, such as septic shock, hemorrhagic shock, or thrombotic events, blood flow needs to be tightly controlled to minimize the incidence of ischemic tissue. These data suggest that the use of volatile anesthetic agents in cases involving such clinical syndromes would not compromise this accurate distribution of blood flow, assuming the physiologic mediators of the distribution are intact. If muscarinic receptor activation is found to be important in metabolic control of blood flow, as recently suggested,[43] then although volatile anesthetic agents may effect this regulation, it is not because of alterations in cell-cell communication.

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