Bronchial vasodilation evoked by increased lower airway osmolarity in dogs

MICHAEL P. ZIMMERMAN AND THOMAS E. PISARRI
Department of Biomedical Sciences, Creighton University
School of Medicine, Omaha, Nebraska 68178-0405

Zimmerman, Michael P., and Thomas E. Pisarri. Bronchial vasodilation evoked by increased lower airway osmolarity in dogs. J. Appl. Physiol. 88: 425–432, 2000.—Hyperosmotic saline solutions stimulate lower airway sensory nerves. To determine whether airway hyperosmolarity evokes neurally mediated changes in bronchial artery blood flow (Qbr), we measured the effect of injection of small volumes (1 ml) of hyperosmotic saline into a right lobar bronchus on Qbr of anesthetized, artificially ventilated dogs. In 14 dogs, hyperosmotic saline (1,200 and 2,400 mmol/l) increased Qbr by 58±12 (SE) and 118±12%, respectively, from a baseline of 8±2 ml/min. Qbr increased within 6–8 s of the injections, peaked at 20 s, and returned to control over 2–3 min. Isosmotic saline had minimal effects. In contrast, hyperosmotic saline decreased flow in an intercostal artery that did not supply the airways. The bronchial vasodilation was decreased by 72±11% after combined blockade of α-adrenoceptors and muscarinic cholinergic receptors and by 66±6% when the cervical vagus nerves were cooled to 0°C. Blockade of H1 and H2 histamine receptors did not reduce the nonvagal response. We conclude that hyperosmolarity of the lower airways evokes bronchial vasodilation by both a centrally mediated reflex that includes cholinergic and adrenergic efferent pathways and by unidentified local mechanisms.

Bronchial artery; hypertonic saline; vagus nerve; autonomic pathways; airway defense reflex; exercise-induced asthma during dry air hyperventilation in dogs, that changes in airway osmolarity could cause reflex bronchial vasodilation by stimulating airway receptors. However, Godden et al. (13) subsequently reported that increases in lower airway osmolarity had no effect on bronchial blood flow. The results of several studies led us to reexamine the effect of lower airway hyperosmolarity on bronchial blood flow. Injection of nonisosmotic solutions into the lower airways stimulates unmyelinated (C-fiber) afferents as well as rapidly adapting (irritant) receptors and evokes well-described elements of the pulmonary chemoreflex: apnea followed by rapid shallow breathing, airway smooth muscle contraction, bradycardia, and systemic hypotension (28). This observation suggested that hyperosmolarity should increase airway blood flow, because stimulation of airway C-fiber afferents with capsaicin increases bronchial blood flow and vascular conductance (6, 26). Similarly, lower airway hyperosmolarity, produced by injection of a small volume of water into a lobar bronchus, evokes reflex bronchial vasodilation (25). In this study, we attempted to determine whether the defense response evoked by hyperosmotic fluid in the lower airway includes bronchial vasodilation. After demonstrating hyperosmotic-evoked bronchial vasodilation, we examined the afferent and efferent pathways of the vasodilation by blocking neural conduction in the cervical vagus nerve and by pharmacological blockade of α-adrenergic and cholinergic autonomic pathways. In addition, because release of histamine from airway mast cells has been hypothesized as the principal mediator of hyperosmotic saline-induced airway obstruction (2), we examined the role of histamine receptors in hyperosmotic bronchial vasodilation.

AIRWAY DEFENSE RESPONSES INCLUDE neural and nonneural mechanisms activated when foreign substances enter the airways. Nonisosmotic fluids evoke defense responses that include cough, changes in breathing pattern, tracheal submucosal gland secretion, and, in asthmatic individuals, bronchoconstriction (2, 10, 17, 21, 23). The tracheal vasculature participates in these defense responses, with tracheal vascular resistance decreasing and vascular permeability increasing with hyperosmolarity of the tracheal lumen (8, 31, 32). The participation of the bronchial circulation in defense responses is of particular interest because change in airway osmolarity may initiate airway narrowing during exercise-induced asthma (1), and bronchial mucosal hyperemia may contribute to the airway narrowing (19). Baile et al. (3) suggested, on the basis of their observation of an increase in bronchial blood flow during dry air hyperventilation in dogs, that changes in airway osmolarity could cause reflex bronchial vasodilation by stimulating airway receptors. However, Godden et al. (13) subsequently reported that increases in lower airway osmolarity had no effect on bronchial blood flow.

METHODS

General. Dogs (18–35 kg) were given acepromazine maleate (1 mg/kg im; PromAce, Aveco); 30 min later they were anesthetized with α-chloralose (60 mg/kg iv). Supplemental doses of α-chloralose (10 mg/kg iv) were given hourly to maintain anesthesia. The trachea was cannulated low in the neck. A catheter (PE-90) was inserted through a rubber stopper in the tracheal cannula and its tip directed into a right lobar bronchus. The chest was opened through the right fifth intercostal space, and the lungs were ventilated with 50% O2 in air by a Harvard respirator (model 613), the expiratory outlet of which was placed under 3–5 cm of water. Tidal volume was set at ~15 ml/kg and ventilation frequency at 15–20 cycles/min. Tidal CO2 was monitored by a Normocap 200 gas analyzer, and end-tidal PCO2 was kept at ~35 Torr by

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

http://www.jap.org 8750-7587/00 $5.00 Copyright © 2000 the American Physiological Society 425
adjusting ventilator frequency. Arterial blood samples were withdrawn periodically, and blood-gas and acid-base values were determined with an automatic analyzer (model 178, Corning); base deficit was corrected by intravenous administration of sodium bicarbonate solution.

Tracheal pressure was measured from a sidearm of the tracheal cannula by a Statham PR23-GG-300 strain gauge. Arterial blood pressure was recorded from a catheter introduced through the right external jugular vein. An electrocardiogram (lead II) was recorded and used by a cardiotachometer (model 7P4C, Grass) to calculate heart rate. The signals representing tidal CO2, tracheal pressure, systemic arterial and left or right atrial blood pressure, heart rate, and blood flow in the bronchial artery (see Measurement of bronchial arterial flow) were recorded by a Grass polygraph (model 7).

Measurement of bronchial arterial flow. The right bronchial (bronchoesophageal) artery and the aortic intercostal artery from which it arose were identified. The right bronchial artery supplies the carina and airways of the right lung; 30% of its flow is to adjacent mediastinal tissues (14). To reduce flow to nonrespiratory tissues, branches supplying the esophagus were ligated. A fine polyethylene catheter was inserted in the intercostal artery distal to the origin of the bronchial artery and advanced retrogradely until its tip was just downstream to the origin of the bronchial artery. With the intercostal artery ligated at the point of cannula insertion, solutions infused slowly through the catheter entered the bronchial circulation. The identity of the bronchial artery was confirmed and its vascular territory outlined by injection of indocyanine green dye (Sigma Chemical) into the intercostal catheter. The bronchial artery and, in some experiments, an adjacent (usually caudal) intercostal artery that did not supply the airways were dissected free of connective tissue. A 1-mm-ID ultrasonic transit-time flow probe (model 1R, Transonic Systems, Ithaca, NY) was placed on the bronchial artery and a 2-mm-ID flow probe (model 2S) on the adjacent intercostal artery. Blood flows in the two arteries were measured by a Transonic T206 small-animal, two-channel ultrasonic flowmeter. Signals representing bronchial and systemic arterial blood pressure were fed to an electronic divider that calculated bronchial vascular conductance as bronchial blood flow per 100 mmHg mean systemic arterial pressure. Because changes in right atrial pressure were never more than 1 mmHg, they were not subtracted from mean arterial pressure in the estimate of conductance. Flow and conductance were recorded by the polygraph.

Injection of solutions into the lower airways. Saline solutions of four different osmolarities were kept in a water bath heated to 37°C: 2,400 (7.2%), 1,200 (3.6%), 300 (0.9%, isosmotic), and 0 (water) mmol/l. Immediately before injection, a small volume (generally 1 ml more than the dead space of the catheter) was withdrawn through a syringe. The solutions were injected rapidly into the catheter that had been passed through the tracheal cannula. Three minutes after the injection, the solution remaining in the catheter dead space was withdrawn, and the lung was hyperinflated to restore compliance.

Blockade of vagal pathways. In some experiments we blocked conduction in the vagus nerves by cooling them to 0°C. For this purpose, the vagosympathetic trunks were dissected free in the midcervical region and loose threads placed around them. The nerves were then placed on the platforms of cooling devices through which alcohol of different temperatures was circulated (15).

Protocol. We measured the changes in blood flow in the bronchial and intercostal arteries evoked by injection of 1–2 ml of solutions of hyperosmotic saline (1,200 or 2,400 mmol/l). We compared these effects with those produced by injection of the same volume of isosmotic (300 mmol/l) NaCl solution or of water. We then repeated the hyperosmotic injection with the vagus nerve cooled to 0°C and finally with the vagus nerves rewarmed.

Pharmacological blockade of autonomic receptors. In some experiments we blocked muscarinic cholinergic receptors and a-adrenoceptors. Postganglionic, parasympathetic (muscarinic) receptors were blocked with atropine sulfate (1 mg/kg iv). The a-adrenoceptors accessible from the bronchial circulation were blocked by infusion of phentolamine mesylate (Regitine, CIBA Pharmaceutical) into the bronchial artery (500 µg/min for 5 min in a volume of 0.5 ml/min). This infusion rate abolishes the bronchial vasconstrictor response to electrical stimulation of the right sympathetic chain (29). Because the action of phentolamine is of short duration, responses after a-adrenoceptor blockade were measured during ongoing phentolamine infusion. Control responses were measured during infusion of vehicle (saline) at the same rate. We measured the effect of atropine alone (9 dogs), phentolamine alone (9 dogs), and atropine and phentolamine together (5 dogs). With two exceptions, different dogs were used in these protocols.

Blockade of histamine receptors. In six dogs, we measured the effect of histamine-receptor blockade on the response to hyperosmotic saline injection after vagotomy. Histamine receptors were blocked by intravenous injection of the H1-receptor-antagonist chlorpheniramine maleate (10 mg/kg; Sigma Chemical) and the H2-receptor-antagonist cimetidine HCl (5 mg/kg; SmithKline Beecham). The postblockade response to hyperosmotic saline was measured 5 min after administration of the histamine-receptor antagonists.

Aerosol delivery of hyperosmotic saline. In eight dogs with intact neural pathways, we measured the vasodilator response to aerosol delivery of 7.2% NaCl solution for 90–150 s. Aerosol of hyperosmotic or isosmotic saline was delivered to the lower respiratory tract by passing the inspiratory flow from the ventilator pump through an ultrasonic nebulizer (model 35 B, DeVilbiss) that generated the aerosol. The nebulizer delivered −0.7 ml/min into the inspiratory line.

Analysis of results. As an index of vasodilation, we calculated bronchial vascular conductance and intercostal vascular conductance as blood flow (milliliters per minute) per 100 mmHg mean arterial pressure. Changes in bronchial arterial blood flow, intercostal arterial blood flow, and bronchial and intercostal vascular conductance were calculated by comparing the control value before the injection of the osmotic stimulus with the value at the point of maximal change. Control measurements were averaged over 30 s immediately before the stimulus. The peak response was averaged over the 5–s period after the stimulus in which the change from control was maximal; this invariably occurred 15–30 s after the stimulus. However, the change was recorded as zero if it was not greater than the maximal spontaneous variation of flow in the 2 min preceding the injection. The time course of the response was plotted by averaging the response in 5-s intervals for 120 s after the stimulus. Data were sampled at 200 Hz by a MacLab/8/s data-acquisition system (ADInstruments) and averaged by using Chart software.

Results are expressed as means ± SE. Statistical analyses of the effect of receptor antagonists or vagal cooling and rewarmed on changes in flow, conductance, arterial pressure, and heart rate were made by using ANOVA for repeated
measures. If a significant effect was detected, individual means were compared by constructing contrasts using Super-ANOVA statistical software. Statistical significance was accepted if \(P < 0.05\).

RESULTS

In 14 dogs, baseline flow in the right bronchial (bronchoesophageal) artery was 8.1 ± 1.4 ml/min (range 2–20 ml/min). Mean arterial pressure was 111 ± 3 mmHg (range 87–132 mmHg), and baseline bronchial vascular conductance was 7.1 ± 1.3 ml·min\(^{-1}\)·100 mmHg\(^{-1}\) (range 2.3–14.5 ml·min\(^{-1}\)·100 mmHg\(^{-1}\)).

Effects of osmolality on bronchial vascular conductance. Injection of a small bolus (1 ml) of hyperosmotic (7.2%, 2,400 mmol/l) NaCl into a right lobar bronchus increased bronchial artery flow to 17.3 ± 3.2 ml/min and decreased heart rate (from 145 ± 8 to 108 ± 7 beats/min) and mean arterial pressure (from 111 ± 3 to 96 ± 5 mmHg) (Fig. 1). Bronchial vascular conductance began to increase within 5 s of the injection, reaching a peak of 16.7 ± 2.9 ml·min\(^{-1}\)·100 mmHg\(^{-1}\) 15–20 s after injection and returning to the control level after 2–3 min. Isosmotic saline had no significant effect (Figs. 1 and 2).

The vasodilator response to hyperosmotic saline was concentration dependent; the increase in bronchial vascular conductance in response to 3.6% NaCl (1,200 mmol/l) was smaller than that in response to 7.2% (\(P < 0.01\)). The response to 7.2% was similar to the response to water (Figs. 1 and 2).

To determine whether the vasodilator response to hyperosmotic saline was specific to the airway vasculature, we compared the bronchial vasculature with nonairway vasculature supplied by an intercostal artery that did not have bronchial branches. Intercostal blood flow fell or was unaffected by hyperosmotic sodium chloride (0.9%) had no effect. The intercostal vascular conductance was 7.1 ± 1.3 ml·min\(^{-1}\)·100 mmHg\(^{-1}\) (range 2.3 to 6.5 ml·min\(^{-1}\)·100 mmHg\(^{-1}\); controls). Injection of a small bolus (1 ml) of hyperosmotic saline (7.2% NaCl) into a right lobar bronchus increased bronchial artery flow to 17.3 ± 3.2 ml/min and decreased heart rate (from 145 ± 8 to 108 ± 7 beats/min) and mean arterial pressure (from 111 ± 3 to 96 ± 5 mmHg) (Fig. 1). Bronchial vascular conductance began to increase within 5 s of the injection, reaching a peak of 16.7 ± 2.9 ml·min\(^{-1}\)·100 mmHg\(^{-1}\) 15–20 s after injection and returning to the control level after 2–3 min. Isosmotic saline had no significant effect (Figs. 1 and 2).

The vasodilator response to hyperosmotic saline was concentration dependent; the increase in bronchial vascular conductance in response to 3.6% NaCl (1,200 mmol/l) was smaller than that in response to 7.2% (\(P < 0.01\)). The response to 7.2% was similar to the response to water (Figs. 1 and 2).

To determine whether the vasodilator response to hyperosmotic saline was specific to the airway vasculature, we compared the bronchial vasculature with nonairway vasculature supplied by an intercostal artery that did not have bronchial branches. Intercostal blood flow fell or was unaffected by hyperosmotic saline; the intercostal vascular conductance was unchanged (Figs. 1 and 3).

Blockade of vagal conduction. To examine the involvement of vagal pathways in the response to hyperosmotic solution, we interrupted neural conduction in the cervical vagus nerves by cooling them to 0°C. Blockade of vagal conduction resulted in small increases in arterial pressure in most of the dogs, and reduced bronchial artery flow and bronchial vascular conductance in each of 14 dogs, conductance falling from 8.8 ± 1.3 to 6.5 ± 0.9 ml·min\(^{-1}\)·100 mmHg\(^{-1}\) (\(P = 0.05\)). With the vagus cooled, injection of 7.2% NaCl still significantly increased bronchial vascular conductance, but the increase was reduced to 33 ± 6% of the intact response (Fig. 4; \(P < 0.001\)). In only three of the dogs was the response completely abolished by vagal cooling; in the others, the response during cooling ranged from 21 to 74% of the intact response. Rewarming the vagus restored the original response.

Blockade of cholinergic receptors. To determine the autonomic pathways that mediate the vasodilation, we compared the response to hyperosmotic saline before and after blockade of cholinergic pathways (with atropine, 1 mg/kg iv) in nine dogs. Atropine increased the resting heart rate from 93 ± 6 to 118 ± 14 beats/min in these dogs and abolished the decrease in heart rate in response to hyperosmotic saline. Atropine caused small and variable changes in baseline bronchial vascular conductance but reduced the peak vasodilation to hypertonic saline by 50 ± 6% (\(P < 0.001\); Fig. 5). After atropine, the vasodilation to hypertonic saline was slower, and the peak vascular conductance was reached 20–25 s after the stimulus. –5 s later than with cholinergic pathways intact.

Blockade of bronchial vascular a-adrenoceptors. In nine dogs with intact cholinergic pathways, we compared the response to hyperosmotic saline before and during infusion of the a-adrenoceptor antagonist phentolamine. Infusion of phentolamine locally into the bronchial artery had no effect on resting mean arterial pressure or on the small fall in arterial pressure in
response to hyperosmotic saline. Phentolamine did not change resting bronchial vascular conductance but reduced the peak vasodilation to hypertonic saline by 58 ± 6% (P < 0.05; Fig. 6).

Combined cholinergic and α-adrenergic blockade. The results of separate cholinergic and α-adrenergic blockade suggested that, if the two effects were additive, the vasodilation might be entirely accounted for by these two pathways. To examine this possibility, we measured the effect of combined cholinergic and α-adrenergic blockade in five dogs. Combined blockade had no consistent effect on baseline bronchial vascular conductance (2.8 ± 1.0 and 3.2 ± 1.1 ml·min⁻¹·100 mmHg⁻¹ before and after blockade, respectively). In these dogs, the combined blockade reduced the peak vasodilation to hypertonic saline by 72 ± 11%, reducing the change in bronchial vascular conductance from 6.8 ± 1.8 to 2.1 ± 1.3 ml·min⁻¹·100 mmHg⁻¹ (P < 0.01); however, there was still significant vasodilation in the presence of the combined blockade (P < 0.05).

Blockade of histamine receptors. In six dogs, we examined the role of histamine in the nonvagal component of the vasodilation to hyperosmotic saline. After cervical vagotomy, injection of 7.2% NaCl solution increased bronchial vascular conductance by 4.2 ± 1.1 ml·min⁻¹·100 mmHg⁻¹. We then blocked both H₁ and H₂ histamine receptors with intravenous infusion of chlorpheniramine and cimetidine, respectively. The histamine-receptor blockade had no effect on the response to hyperosmotic saline; bronchial vascular conductance increased by 4.3 ± 1.2 ml·min⁻¹·100 mmHg⁻¹ in the presence of blockade (Fig. 7).

Hyperosmotic aerosol. Eight animals with intact neural pathways were ventilated with 7.2% NaCl aerosol for 90–150 s. Ventilation with hyperosmotic aerosol had no effect on mean arterial pressure. Bronchial vascular conductance rose in seven of the dogs (Fig. 8), increasing from 10.3 ± 1.8 to 12.8 ± 2.2 ml·min⁻¹·100 mmHg⁻¹ (P < 0.05) for the group as a whole. The vasodilation began 10–20 s after the onset of aerosol exposure, and bronchial vascular conductance returned to baseline within 3 min of the termination of the aerosol. Isosmotic saline aerosol, tested in five of the dogs,
DISCUSSION

The results of this study indicate that the presence of a small volume of nonisomotic fluid in the lower airways activates airway responses that include bronchial vasodilation. The vasodilation was manifest as an increase in bronchial artery flow in the face of a simultaneous decrease in systemic arterial pressure. The response was not part of a general systemic vasodilation: flow decreased in an adjacent intercostal artery that did not supply the airway, suggesting a specific vasodilation of the circulation to the airways. The demonstration of hyperosmotic-evoked bronchial vasodilation extends our previous finding that hyposmotic solutions cause bronchial vasodilation, whereas solutions isosmotic to the normal airway lining fluid have no effect (25).

These findings are in contrast to those reported by Godden et al. (13), who found no changes in tracheal or bronchial blood flow, measured by microsphere injection, after 3, 12, or 25 min of ventilation of dogs with hyperosmotic saline aerosol. Differences in the method of delivery of the hyperosmotic solution do not completely account for the difference. Hyperosmotic aerosol in our studies evoked bronchial vasodilation within 20 s in all but one of the dogs, although the increase in vascular conductance was small compared with the delivery of similar volumes by direct injection. The absence of detectable vasodilation in the study of Godden et al. may be due in part to the relatively milder stimulus (5% NaCl), the lower rate of aerosol delivery in most of their experiments, and the longer time of exposure to the stimulus before flow measurement. The greater effectiveness of directly injected solution than of aerosol of the same osmolarity is probably due to the more rapid and concentrated delivery of the hyperosmotic solution.
stimulus to the airway sensory endings. Our results demonstrate that a sufficient change in osmolarity evokes a rapid bronchial vasodilation that, on average, doubles bronchial blood flow.

Osmolarity-sensing mechanisms in the lower airway. Changes in osmolarity in the airway might be detected by sensory nerve endings, by cells of the immune system, or directly by the vascular smooth muscle of the bronchial arterioles. Our results suggest that much of this bronchial vasodilation is initiated by osmolarity-sensitive vagal afferent fibers. Cooling the cervical vagus nerves, which eliminates afferent and efferent vagal pathways but preserves local mechanisms (including axon reflex pathways), eliminated two-thirds of the vasodilation. Thus the majority of the vasodilation in dogs is due to centrally mediated neural reflexes that include afferent and/or efferent pathways in the vagus nerves. Injection of either water or hyperosmotic solution into a lobar bronchus stimulates the endings of two classes of sensory nerves of the airway: unmyelinated (C-fiber) afferents and rapidly adapting pulmonary stretch receptors (or “irritant receptors”) (28). This stimulus also evokes reflex changes in breathing, airway smooth muscle tension, and arterial pressure that resemble the pulmonary chemoreflex evoked by stimulation of pulmonary and airway sensory C fibers (7). Indeed, selective stimulation of sensory C fibers with capsaicin evokes a bronchial vasodilation of magnitude and time course similar to that evoked by hyperosmotic saline in the present experiments (6, 26). It is, therefore, likely that the osmolarity-evoked bronchial vasodilation is mediated, at least in part, through the same pathways.

However, the population of afferent fibers activated by the hyperosmotic stimulus is not identical to that activated by injection of capsaicin intravascularly. The hyperosmotic stimulus is confined to sensory endings in the intrathoracic airways but activates both C-fiber and rapidly adapting-receptor endings (28), whereas capsaicin activates C-fibers endings accessible from the entire vasculature but has little effect on myelinated afferents. This difference is reflected in the relative effects of the two stimuli: the effects of capsaicin (26) and hyperosmotic saline (present study) on the bronchial circulation were similar, but the effects of capsaicin on heart rate and systemic arterial pressure were nearly twice those of hyperosmotic saline. Stimulation of rapidly adapting receptors by hyperosmotic saline could conceivably contribute to the bronchial vasodilation evoked by this stimulus. There is circumstantial evidence that these afferents contribute to bronchial vasodilation evoked by cooling the airways (27) or by injecting water into the airways (25). However, in the absence of a selective stimulus to rapidly adapting receptor endings, the effects of these afferents on the bronchial circulation cannot be established.

Neural vasodilator mechanisms. The bronchial vasculature is influenced by both adrenergic sympathetic and cholinergic and noncholinergic parasympathetic efferents (5, 12, 30). Much of the hyperosmotic saline-evoked vasodilation was mediated by cholinergic pathways, because atropine reduced the response by 50%. Thus the role of parasympathetic cholinergic pathways in the response to hyperosmotic saline is similar to that in reflex bronchial vasodilation evoked by capsaicin (6, 26) or water (25).

The reduced vasodilation after α-adrenoceptor blockade with phentolamine suggests that sympathetic pathways are also important in the bronchial vasodilation. Sympathetic pathways participate in the bronchial vasodilation to carotid sinus nerve stimulation (20), capsaicin (6), and water (25). Nonetheless, the effect of α-adrenoceptor blockade is puzzling because infusion of phentolamine into the bronchial artery had no effect on baseline bronchial vascular conductance. In previous studies, α-adrenoceptor blockade has been found to increase bronchial artery flow in awake dogs (14) but not in anesthetized dogs (4, 25). If the hyperosmotic-evoked vasodilation involved a withdrawal of sympathetic constrictor tone, we should have seen an increase in conductance with interruption of these pathways. One possible explanation is suggested by our observation in a few experiments of a transient increase in flow at the beginning of phentolamine administration, followed by a rapid return to the prephentolamine level. This suggests that the sympathetic constrictor tone is replaced by other constrictor mechanisms, perhaps local metabolic factors, that restore flow to the level appropriate to the needs of the vascular bed. Unlike sympathetic tone, these local factors could not be withdrawn as part of the response to a hyperosmotic stimulus. Consequently, the vasodilation to a hyperosmotic stimulus would be blunt even in the absence of change in baseline tone.

Our results also suggest an interaction between the sympathetic constrictor pathways and parasympathetic vasodilator pathways. In the bronchial vasculature of dogs, sympathetic pathways inhibit vagally mediated vasodilation (18). If so, sympathetic vasoconstriction, and consequently the vasodilator effects of withdrawal of sympathetic discharge, would be attenuated after cholinergic blockade. Such interaction is suggested by the less than additive nature of the separate cholinergic and α-adrenergic pathways. Thus, whereas atropine reduced the hyperosmotic vasodilation by 50%, and phentolamine reduced it by 58%, combined blockade reduced the response by only 72%.

Although the combined α-adrenoceptor- and cholinergic-mediated responses account for all of the vagally mediated influence on the peak flow response, our data do not rule out a role for autonomic cotransmitters such as vasointestinal peptide. This neurotransmitter has been suggested to account for nonadrenergic, noncholinergic baroreflex bronchial vasodilation in awake dogs (14). During electrical stimulation of the peripheral vagus nerve in dogs, rapid onset vasodilation is atropine-sensitive, but noncholinergic hexamethonium-sensitive vasodilation develops more slowly (30). Thus, whereas the peak response to hyperosmotic saline might be accounted for entirely by cholinergic and adrenergic mechanisms, noncholinergic autonomic
neurotransmitters might participate in the later phases of the vasodilation.

Although the qualitative similarities between the response to hyperosmotic saline in dogs and asthmatic humans suggest similar neural pathways, the role of the canine airway vasculature in thermoregulation raises the possibility that bronchial vascular control is not identical. The canine bronchial vasculature, unlike that of humans, supplies heat and water for conductive and evaporative heat loss during panting. It could be speculated that the canine airway sensitivity to osmolarity changes serves a regulatory role in balancing fluid loss to airway blood flow as part of a thermoregulatory reflex. However, the observation that the response to osmolarity changes in dogs includes bronchoconstriction and bradycardia (28) argues against a thermoregulatory role.

Nonreflex vasodilator mechanisms. About 30% of the hyperosmotic saline evoked vasodilation was mediated by pathways that did not require vagal communication to the central nervous system. This is in contrast to the vasodilation evoked by water, which in most dogs is mediated entirely by central vagal pathways (25). The nonvaginal pathways could include release of mediators from osmolarity-sensitive cells of the immune system (1) or release of neuropeptides from the terminals of airway C fibers (5).

Histamine is a bronchial vasodilator (16) and is released from mast cells by hyperosmotic solutions in vitro (9). However, in the present study, combined blockade of H1 and H2 receptors did not change the vasodilation to hyperosmotic saline in vagotomized dogs. Thus our results indicate that histamine is not necessary for the nonvagal component of the vasodilation. This does not exclude the possible involvement of other mast cell mediators (22).

We did not directly examine the contribution to the hyperosmotic vasodilation of neuropeptides released from the endings of sensory C fibers. Although sensory neuropeptides play an important role in the airways of rodents, evidence of their importance in the airways of humans and other larger mammals is weak (5, 33). However, electrical stimulation of the peripheral vagus nerve of dogs causes some bronchial vasodilation after ganglionic blockade with hexamethonium (30). This vasodilation is reduced by infusion of a specific antagonist of the receptor for the sensory neuropeptide calcitonin gene-related peptide (24). Moreover, bronchial vasodilation to capsaicin is not entirely eliminated by vagotomy in some dogs, and the persisting vasodilation is marginally enhanced during infusion of an inhibitor of endogenous neuropeptide-degrading enzymes (26). This evidence suggests the potential for sensory neuropeptide release when airway C fibers are stimulated by hyperosmotic saline.

Relationship to exercise-induced airway obstruction. An increase in osmolarity of the airway surface liquid and submucosa subsequent to airway drying has been hypothesized as the stimulus to bronchial vasodilation during exercise (1). The change in osmolarity that might occur during exercise or hyperpnea is uncertain, but it is unlikely to be as large as that produced by the bolus injections we used. Although the osmotic stimulus during exercise would be spread throughout the airways, and thus activate more endings than our localized injection, it is nonetheless reasonable to assume that our stimulus was greater than might be expected during exercise or other hyperpnea. However, our experiments were conducted in nonsensitized mongrel dogs. The sensitivity of these dogs to the osmotic stimulus would be expected to be low, as is the case with nonasthmatic humans. In hypersensitive individuals, smaller changes in osmolarity might be sufficient to activate these same responses. Hyperpnea, particularly of cold dry air, may activate additional defense responses by cooling the airway. Both cooling and hyperosmolarity activate pathways that produce bronchoconstriction and vasodilation (present results, Refs. 27 and 28). Whereas the cold stimulus alters airway stretch receptor, but not airway C-fiber discharge (11), the hyperosmotic stimulus activates airway C fibers and rapidly adapting receptors (28). During cold dry air hyperpnea, the activation of several neural pathways could sum to produce airway and vascular responses that contribute to obstruction.

Hyperventilation with dry air, which is believed to mimic the stimulus to exercise-induced asthma, increases bronchial blood flow in artificially ventilated dogs (3). However, neither vagotomy nor α- or β-adrenoceptor blockade attenuated dry air ventilation evoked bronchial vasodilation (4), in contrast to the hyperosmolarity evoked vasodilation in the present experiments. This would seem to suggest that hyperventilation-induced bronchial vasodilation is mediated by a pathway different from the largely neural mechanism of hyperosmotic-induced vasodilation. However, in the dry-air ventilation experiments (3), airway blood flow was measured only after 25 min of hyperventilation, whereas we measured immediate responses to the hyperosmotic stimulus. It is possible that the balance between local and neurally mediated vasodilation changes during an extended hyperosmotic stimulus.

In summary, we have shown that a change in osmolarity in the lower airway initiates a centrally mediated bronchial vasodilation that could contribute to the airway obstruction of exercise-induced asthma.

We thank Douglas Lassiter, Albert Dangel, and Ronald Brown for technical assistance.

This investigation was supported by American Heart Association Grant-in-Aid 95008270, Council for Tobacco Research Scholar Award SA027, and the State of Nebraska Cancer and Smoking Related Disease Program (LB 595).

Address for reprint requests and other correspondence: T. E. Pisarri, Dept. of Biomedical Sciences, Creighton Univ. School of Medicine, 2500 California Plaza, Omaha, NE 68178-0403 (E-mail: tpisarri@creighton.edu).

Received 10 July 1999; accepted in final form 28 September 1999.

REFERENCES


432 HYPEROSMOLARITY-EVOKED BRONCHIAL VASODILATION


