tropic antibody and antigen. The mechanism by which eosinophils might act to increase the level of histamine following this interaction is obscure. Also, the conclusion that the eosinophil itself alters the level of histamine rests upon the assumption that AES itself does not directly alter mediator release from mast cells and basophils nor alter the degradation of mediators. Although AES preferentially ablates eosinophils from the blood and tissues of the guinea pig, we cannot exclude the possibility of an effect on the membranes of mast cells and basophils which might alter their ability to release histamine by a heterocytotropic antibody-antigen interaction. Against such an interpretation is the finding that AES is unable to release histamine following direct injection into the peritoneal cavity (results not shown). Nonetheless, administration of AES in some experiments does result in a blunting of the increase of basophils seen after administration of rabbit serum. This emerges from the statistical analyses of the basophil levels in the animals shown in Figure 1.

Our result, showing that ablation of the eosinophil reduces the level of histamine, is at variance with the reports of Hubsercher,6,9 who demonstrated an increase in histamine release in the absence of eosinophils. However, his data were derived from experiments with human peripheral blood leukocytes sensitized with IgE antibody. Our result is also directly opposed to that predicted by Goetzl et al10 who propose that eosinophils dampen the level of mediators by their ability to enzymatically degrade the mediators. Clearly, further studies of the eosinophil are needed to define its role in immediate-type hypersensitivity reactions.

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DISCUSSION

Dr. Stechschulte: Does your antisera alter the function of the neutrophil?
Dr. Gleich: The expression of the Arthus reaction was not altered. If that reaction is indeed mediated by the neutrophil, then the antisera did not alter neutrophil function in that situation.

Vasoactive Mediators in the Pathogenesis of Experimental Acute Respiratory Failure*

Roger C. Bone, M.D., F.C.C.P.; Scott Lerner, M.D.; Daniel Stechschulte, M.D.; Everett Murphy, M.D.; John Wolfe, M.D.; Richard Sobonya, M.D.; and Jerry Hood, M.D.

Acute respiratory failure is a common and often lethal lung reaction that occurs from many causes. Therapy at the present time is supportive. The mechanism of injury and the cause for increased alveolar-capillary permeability is unknown.

In an anesthetized canine model of acute respiratory failure produced by the intratracheal instillation of hydrochloric acid, we found an elevated histamine concentration in intratracheal secretions and elevated arterial histamine levels. An additional smooth muscle contracting substance was found in the intratracheal secretions with characteristics of slow-reacting substance of anaphylaxis (SRS-A). These vasoactive mediators may be important in the injury reaction of this model of acute respiratory failure.

METHODS

In 18 dogs, 3 ml/kg of 0.1N hydrochloric acid was instilled into the tracheobronchial tree after endotracheal intubation. Three ml/kg saline solution was similarly instilled into 11 control animals. The animals were anesthetized with 30 mg/kg thiopental sodium and paralyzed with 2 mg/kg succinylcholine given intravenously. Muscle relaxation and anesthesia were maintained with a continuous succinylcholine drip at 30 mg/hr and thiotal sodium 50 mg/hr. The animals were ventilated by a Bennett MA-1 ventilator with room air. A flow-directed pulmonary artery catheter was placed in each animal to monitor pulmonary capillary wedge pressure. Arterial blood gases and static compliance of the lungs and chest wall were measured.

Arterial histamine levels were measured after thiopental administration, after institution of mechanical ventilation, after succinylcholine administration and 1, 10, 60, 120, and 180 minutes after instillation of acid or saline solution. Histamine was measured by the enzymatic isotopic assay described by Snyder and colleagues2 and modified by Beaven and coauthors.3 The basis of the assay is the enzymatic

*From the Department of Medicine, University of Kansas Medical Center, Kansas City. Reprint requests: Dr. Bone, University of Arkansas Medical Center, Little Rock 72201

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conversion of histamine to form methylhistamine. S-adenosyl methionine with "C-labeled is used as the methyl donor, and guinea pig brain histamine N-methyl transferase as the enzyme.

The smooth muscle contracting substance was assayed with guinea pig terminal ileum suspended in oxygenated Tyrode's solution at 37°C. Contractile activity was measured in the presence of atropine at a concentration of 10^{-4}M and mepyramine maleate at a concentration of 10^{-6}M.

All measurements are expressed as mean ± standard error. Statistical analysis was performed by Student's t test or Chi square analysis. The probability of an observation occurring by chance alone less than 5 percent of the time (P < 0.05) was considered significant.

RESULTS

The arterial oxygen tensions (PaO_{2}) and effective static compliances were significantly less in the acid-injured animals compared to saline control animals (Fig 1). The arterial pH was less in the acid injured animals (Fig 1). The pulmonary capillary wedge pressures were not different in the acid-injured animals (8.4 ± 1 mm Hg) compared to saline control animals (8.1 ± 1 mm Hg). Arterial histamine was greater in the acid-injured animals (Fig 2). The histamine levels were not different after anesthesia (1.3 ± 0.2 ng/ml), muscle relaxation (2.1 ± 0.5 ng/ml) and after institution of mechanical ventilation (1.7 ± 0.6 ng/ml) but increased greater than 6 times control after instillation of acid (Fig 2). Arterial histamine decreased to twice control levels by one hour after instillation. The concentration of histamine in the intratracheal secretions were 30.3 ± 3.8 ng/ml.

Tracheal secretions obtained from acid-injured animals at two hours after instillation had a prolonged and delayed smooth muscle contracting substance in 7 of the 11 secretions studied. This smooth muscle contracting substance had characteristics of slow-reacting substance of anaphylaxis (SRS-A). It was ethanol extractable and was unaffected by atropine or mepyramine maleate and was inactivated by arylsulfatase. Secretions obtained two hours after instillation of acid had a pH of 7.4 and a total protein of 5.0 ± 0.2 gm/100 ml which was not different than the total protein of plasma (5.6 ± 0.3 gm/100 ml).

The lung weight/body weight ratio of the acid injured animals (2.20 ± 0.10 × 10^{-2}) were significantly greater than the saline controls (0.85 ± 0.06 × 10^{-2}). A severe hemorrhagic pulmonary edema was seen on histologic examination.

DISCUSSION

Diseases associated with pulmonary edema from increased microvascular permeability could be mediated by endogenously released vasoactive substances. Histamine has been shown to cause systemic exchanging vessels to leak excessively. Pietra et al demonstrated a leakage of carbon particles from bronchial venules following histamine infusion in dogs. These animals had increased lung water on postmortem examination.

**Figure 1.** Change in physiological measurements. The arterial oxygen tension (PaO_{2}), effective static compliance and pH are decreased from baseline measurements. The horizontal axis is expressed in minutes. NS = not significant; SEM = standard error of the mean.

**Figure 2.** Change in plasma histamine before and after acid injury and saline instillation. The brackets represent standard error of the mean. Measurements made after anesthesia, muscle relaxants, and mechanical ventilation were not different in the two groups of animals. At 120 and 180 minutes the histamine levels were not different.
Brigham et al found that histamine infusion caused a dose-related, reversible increase in lung vascular permeability to protein in unanesthetized sheep. He later showed that the H2 receptor inhibitor diphenhydramine infused with histamine phosphate prevented the histamine-induced pulmonary edema. Metiamide, an H3 receptor inhibitor, infused with histamine did not prevent the increased lung vascular permeability. Morphologic studies suggest that histamine causes large gaps to appear between venular endothelial cells, perhaps resulting from endothelial cell contraction.

In contrast to the release of preformed histamine, slow-reacting substances of anaphylaxis (SRS-A) is not present in the cell in the preformed state and the kinetics of its release are different. On a weight basis, it is at least as potent as histamine as a smooth muscle contractor. Its presence in tracheal secretions after acid-induced injury is of particular interest since it has been shown to increase vascular permeability and decrease pulmonary compliance. This vasoactive mediator has been shown to have a prolonged action compared to the transient effect of histamine. Both mediators could synergistically increase microvascular permeability and alter pulmonary physiology.

In summary, our studies show that histamine and a smooth muscle contracting substance are released after acid-induced injury in the dog. These vasoactive mediators may be important in pulmonary edema from increased microvascular permeability. Since antagonists to histamine and SRS-A are available, their true role in experimental increased permeability pulmonary edema can be assessed.

REFERENCES


DISCUSSION

Dr. Petty: Your model is appealing because it has the same degree of compliance abnormality and rapidly developing hypoxemia that is found in humans, but I think that there's one thing missing: the latent period between the injury and the development of this syndrome.

Dr. Bone: We were using such large doses of hydrochloric acid that it produced death in the animals. However, if we use smaller doses, we don't have the lethal reactions, and we may be able to see a latent period.

Dr. Turner-Warwick: With the amounts of histamine and SRS-A which you see in the secretions, would you expect an effect on the airways?

Dr. Bone: Although the predominant abnormality which we see in this syndrome is decrease in compliance, Drazen and others have shown airway resistance changes after mediator infusion. We did have a change in airway resistance, but compliance changes more.

Dr. Lakshminarayan: There seems to be a lag between the rises in the levels of histamine and the physiologic changes.

Dr. Bone: That's correct. The greatest physiologic changes seemed to better parallel the finding of slow-reacting substance than histamine.

Dr. Chester: Hypoxemia alone produces a number of changes in mediator release. Do you have any information whether hypoxemia alone may increase histamine?

Dr. Bone: The saline treated animals were also hypoxic, so hypoxemia alone could not have produced the mediator changes.

Dr. Brody: Dr. Bone, in the animal model using acid, you'd expect a lot of damage to type I and II alveolar lining cells. Can you separate the effects of histamine on vascular permeability from the effects of damaged alveolar epithelial cells? Is this a problem in your model?

Dr. Bone: I don't think that it's a problem. Studies by Pietra showed that histamine increased pores in the bronchial vasculature before the alveolar-capillary membrane. For that reason, people are thinking about changes in the small airways as well as the alveolar-capillary membrane in this syndrome.

Lymphocytic Acid-Hydrolases and Response to Mitogens in Cystic Fibrosis*

Jack Lieberman, M.D., F.C.C.P., and W. Kaneshiro, B.S.

Marchi et al were the first to report that lymphocytes were hypo-responsive to phytohemagglutinin

*From the UCLA San Fernando Valley Medical Program, and the Veterans Administration Hospital, Sepulveda, California. Supported by Grant no. HL 21156 from the U.S. Public Health Service, and the Medical Research Service of the Veterans Administration.

Reprint requests: Dr. Lieberman, VA Hospital, Sepulveda, California 91343

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