CONCLUSIONS
Studies of the structural and mechanical properties of ASM have provided insights into its role in pathophysiologic conditions such as asthma. Perhaps the most significant finding is that shortening capacity is increased in sensitized ASM. This early change precedes any change in isometric force development and is the test-tube analogue to allergic bronchoconstriction. To what extent changes in internal resistance to shortening and in the properties of cytoskeletal proteins are responsible for the changes in shortening capacity remains to be discovered.

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Pathogenesis of Airway Hyperreactivity*
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Increased irritability (hyperreactivity) of the airways to a variety of stimuli is common to most patients suffering from asthma. The mechanisms causing airway hyperreactivity are unknown. Learning about them may be fundamental to an improved understanding of (1) the pathogenesis and course of chronic airway diseases such as asthma, whether allergic or nonallergic; and (2) the mechanisms by which acute airway injury, whether immunologic or nonimmunologic, leads to airway dysfunction. Perhaps current scientific debate over the mechanisms of hyperreactivity may derive from the fact that the causes of hyperreactivity developing in these two settings are distinctly different. There is relatively little information on the mechanisms of the chronic hyperreactivity that occurs in asthmatic patients. Furthermore, an irreconcilable problem in studies concerning the possible mechanisms of hyperreactivity in this chronic disorder is the inability to separate causes from consequences of the disease. Therefore, we shall concentrate in this review on information that may provide insight into the mechanisms by which airway injury leads to acute airway hyperreactivity. We have hypothesized that acute hyperreactivity results from mediators generated, after airway injury, from normal lung constituents, including mucosal cells (and, in particular, neuroepithelial bodies, mast and epithelial cells), which augment neuromuscular responsiveness of the airways. Given the different pathologic features of asthma (Table 1), it is quite possible that other mechanisms are responsible for the chronic hyperreactivity that occurs in this disorder.

To study in more detail the cellular pathophysiology linked to the development of acute hyperreactivity, various investigators have focused on acute experimental airway disorders in animals which are characterized, in part, by hyperreactivity. We found that an exposure as brief as 15 minutes to 3 ppm of ozone dramatically increases muscarinic reactivity 30 minutes after exposure, a change which is as striking as that which occurs two hours after a two-hour exposure to the same ozone concentration. Associated with these physiologic findings are pathologic ones characterized by degenerative changes in ciliated and goblet cells of the mucosa and by a significant increase in mucosal mast cell members. When hyperreactivity first manifests, there appears to be no increase in inflammatory cells, although this becomes quite florid by four days after exposure, when muscarinic reactivity has reverted to normal. This temporal dissociation between acute ozone-induced hyperreactivity and the airway neutrophilic infiltration in guinea pigs is similar to some investigators' findings in dogs with SO2-induced airway hyperreactivity but contrasts with other investigators' experience in dogs with ozone-induced hyperreactivity. Our impressions were corroborated by a subsequent study that showed a similar degree and time course of hyperreactivity in guinea pigs depleted of airway and circulating granulocytes by cyclophosphamide pretreatment. Thus, rather than being a cause of airway hyperreactivity, neutrophilic infiltration of the airways appears to be a consequence of the acute, nonimmunologic airway injury producing the hyperreactivity. Similar studies concerning toluene diisocyanate--induced hyperreactivity in the guinea pig by us and by other investigators have led to the same conclusions. At least in the guinea pig, acute airway hyperreactivity appears to occur by a neutrophil-independent mechanism.

Is the acute increase in reactivity that occurs during an asthma attack neutrophil-related? Perhaps the study most relevant to this question was done by DeMonchy et al, who compared the cellular composition of bronchoalveolar lavage fluid at six to seven hours post-antigen challenge in dual responding asthmatics with lavage findings at the same time after antigen in nonresponders and with findings at two to
three hours post-antigen in dual responding asthmatics. The striking difference in lavage cell composition at six to seven hours between dual responding asthmatics and nonresponders was an increased percentage of eosinophils. This observation has been corroborated by a very similarly designed study reported by Diaz et al. It is interesting that an increase in lavage eosinophils was also found in "stable" allergic asthmatics in two recently published studies by Flint et al and by Kirby et al. In these two studies, it is important to note that the lavage fluid from "stable" asthmatic patients also contained increased numbers of mast cells and/or basophils. Unfortunately, DeMochy et al did not quantitate these cell types. Kirby et al found that increases in these cells and in eosinophils both correlated with airway responsiveness. In none of the aforementioned studies (except perhaps that by Diaz et al) were lavage neutrophil counts increased in asthmatic patients, although Metzger et al found an increase in both neutrophils and eosinophils 48 hours after antigen challenge. Unfortunately, Metzger et al did not evaluate lavage characteristics and the change in reactivity status before and during the first six to eight hours after antigen challenge.

Thus, one piece of evidence which casts doubt on the hypothesis that neutrophilic infiltration causes the acute increase in airway reactivity during an asthma attack is that such attacks are associated with airway eosinophilic (not neutrophilic) infiltration and exudation. Another piece of evidence is that airway reactivity, in fact, increases soon after "the immediate response" in dual responding asthmatics. In our experience, this occurs 45 to 105 minutes after antigen challenge (ie, as soon as we could confidently remeasure reactivity after the immediate response had resolved), when there may be no airway inflammatory cell infiltration and/or exudation, based on our current knowledge. Our subsequent experience has indicated that prednisone pretreatment, which itself did not affect baseline airway resistance or reactivity, abolished this early onset increase in airway reactivity and the late asthmatic response without affecting the immediate response in such dual responding asthmatic patients. Whether components of the ensuing airway inflammatory cell infiltration and exudation maintain the hyperreactive state after immune or nonimmune airway injury remains to be fully investigated.

If this early onset increase in reactivity after immunologic airway injury does not appear to be related to inflammatory cell infiltration of the airways, is it perhaps related to the generation by normal lung cell constituents of mediators that increase reactivity status? At present, there is very little information from studies in vivo of atopic asthmatics concerning this issue. However, published data and studies in vitro of passively sensitized human bronchi and of dispersed human lung mast cells clearly support this notion. Allergensensitized airway tissue, independent of factors that may circulate in vivo, has the inherent capacity to elaborate spasmogens on antigen challenge, which may augment airway reactivity.

Likewise, is the early onset increase in airway reactivity that we and others have observed after experimental nonimmunologic airway injury related to the generation by normal lung cell constituents of mediators that can augment reactivity? Evidence is accumulating from studies in vivo to support this concept. However, it is very unclear at present what the precise mediators are in different experimental disorders. Perhaps an equally or more important subject requiring future investigation is learning the mechanisms by which such mediators augment airway reactivity.

It seems probable to us that the cellular events causing hyperreactivity do so by increasing airway neuromuscular responsiveness, and not by producing tissue inflammatory cell infiltration or edema. In general, this effect at airway neuromuscular junctions may manifest in two ways: (1) prejunctionally, by decreasing the degradation or increasing the release of bronchoconstricting mediators, such as the endogenous neurotransmitter ACh (or, alternatively, by increasing the degradation or decreasing the release of endogenous bronchodilating mediators); and (2) postjunctionally, by increasing the responsiveness of airway smooth muscle itself upon stimulation. Both pre- and postjunctionally, effects on these excitable cells may be mediated by (1) membrane potential- (or voltage)-dependent mechanisms; and/or (2) voltage-independent events. Most, if not all, bronchoconstrictors elicit spasm, at least in part, by cell membrane depolarization. We have recently investigated the effects of increasing extracellular K* on ferret airway muscle responsiveness in vitro to electrical field stimulation or exogenous ACh, where changes in force and cell membrane potential were measured simultaneously in muscle preparations devoid of mucosa and submucosa. This research may be of some relevance to the hyperreactivity occurring after airway injury in that damaged airway cells may leak K* into the microenvironment of airway nerve and smooth muscle cells. K* itself causes airway muscle contraction by cell membrane depolarization. Our results, which were corroborated by parallel studies of airway preparations having (3H)ACh-loaded nerve terminals, strongly suggest that elevations in extracellular K* from 6 to 12 mM augment responsiveness of the airways by increasing the release of endogenous ACh.

In comparison to K*, leukotriene D4, LTD4, appears to have prominent post-synaptic effects on the muscarinic sensitivity of airway smooth muscle. We have found these effects, in part, are due to electromechanical actions of LTD4, on airway smooth muscle. In the presence of 10−10 M LTD4, for 20 minutes, a concentration which depolarizes ferret tracheal muscle but does not cause contraction, ACh concentrations of less than 10−8 M produced greater muscle cell membrane depolarization and associated force generation.
than in the absence of LTD₄ (Fig 1). Leukotriene C₄ also appears to have clear-cut electromechanical effects on airway smooth muscle. In addition, leukotriene E₄ (LTE₄) in other investigators’ experience and leukotriene B₄ (LTB₄) in our own appear to potentiate the sensitivity of guinea pig airway smooth muscle to an exogenous bronchoconstrictor. The mechanisms by which LTE₄ or LTB₄ potentiate airway muscle responsiveness have yet to be fully evaluated.

Thus, it appears to us that the potentiation of one bronchoconstrictor's effect by another at airway neuromuscular junctions may be caused by several mechanisms. It is possible that a final common pathway may be an increase in intracellular Ca²⁺ derived extracellularly through cell membrane Ca²⁺ channels, and intracellularly from membrane- and sarcoplasmic reticulum-bound stores. In future research, it will be important to learn the ways in which Ca²⁺ entry through membrane channels of airway nerve endings and smooth muscle cells may be affected by various mediators and by intracellular second messengers such as the cyclic nucleotides as some of our evidence suggests. New technical advances in cell biology provide the opportunity to further investigate possible cellular and subcellular abnormalities that cause airway hyperreactivity.

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