CHAPTER 10

STRUCTURE–FUNCTION RELATIONSHIPS IN CHRONIC OBSTRUCTIVE PULMONARY DISEASE

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Since flow is the result of a driving pressure (elastic recoil of the lung parenchyma) and of an opposing resistance that constrains flow (obstruction of the airways), a reduction in flow can be due either to a reduced driving pressure or to an increased resistance. In smokers, when pathologic changes involve the small airways they will contribute to airflow limitation by narrowing and obliterating the lumen and therefore increasing the resistance. Active constriction of the airways further compounds the problem. Conversely, when pathologic changes are localized in the lung parenchyma, they will contribute to airflow limitation by reducing the elastic recoil of the lung through parenchymal destruction, as well as by reducing the elastic load applied to the airways through destruction of alveolar attachments, therefore reducing the airway lumen further. Because small airway abnormalities and emphysema coexist in the lungs of smokers, their complex interaction will govern the pathophysiology of chronic obstructive pulmonary disease (COPD).

The present chapter focuses on the relationship between structural abnormalities in small airways and lung parenchyma and pulmonary function in COPD in an attempt to highlight the possible mechanisms contributing to airflow limitation in this disease. Special attention will be paid to the inflammatory component in the lungs of smokers, possibly the key component in the causation of disease. We will see how studies correlating morphology, including inflammation and function, have gone a long way toward improving our understanding of the possible mechanisms by which cigarette smoking might produce COPD.

EVOLUTION OF THE UNDERSTANDING OF COPD

COPD is a very old disease, perhaps one of the oldest lung diseases ever suffered. In 1972, a mummy of an old woman, who had lived in Alaska 1,600 years ago, was discovered. Her lungs showed abundant anthracotic deposits along with areas of centrilobular emphysema (CLE).[1] This is probably the oldest known case of COPD, most likely produced by biomass (open fires) exposure. COPD secondary to exposure to open wood fires while cooking is still an important cause of COPD in many countries[2] and probably has been a cause of COPD ever since fire was introduced for cooking. Most likely, the Alaskan woman coughed, produced sputum, and was dyspneic during exercise. Probably many women were! Similar findings were reported in Egyptian mummies.[1] However, it took many centuries for physicians to understand COPD, and this introduction explains briefly how concepts evolved.

The word "emphysema" is derived from the Greek "to blow into" and therefore is used to mean "air containing" or "inflated." Although the first mention of emphysema dates back to the early seventeenth century, with Bonet (1617) describing a case with "voluminous lungs," progress in our understanding of lung disease due to chronic airflow obstruction has been sporadic and painfully slow.[3] The first description of enlarged airspaces in emphysema in humans, together with illustrations, was provided by Ruysh in 1721.[4] Watson,[5] in 1764, gave a complete anatomic description of a case of emphysema: "The lungs were enormously distended with air . . . this air was extravasate and had burst through the extremities of the bronchi and vesicular substance . . . In a word, the lung was truly emphysematous. In several parts this had formed large bladders . . . The air getting loose in the substance of the lungs cannot be parted with expiration; it subsequently is retained there and the space it occupies prevents as much of the external air being received into the lungs as its own quantity." Morgagni, in his "De sedibus et causis morborum per anatomen indagatis" (1769), described several cases in which the lungs were

turgid, particularly from air.[6] Baillie in 1799[7] and 1807[8] provided a very clear description of emphysema. In his textbook, he states that emphysematous lungs did not collapse at postmortem and filled the pleural cavities and that the alveoli ("cells") were full of air.

The contribution of Laennec in 1826[9] is a crucial one. He recognized that fixation by inflation was essential for the proper examination of the lungs for the presence of disease. He was the first to distinguish between interstitial emphysema and enlarged air sacs, or "emphysema proper." He appreciated that airspaces enlarge with increasing age, and he distinguished these physiologic changes from emphysema. More importantly, he related the structural abnormalities to the clinical syndrome as he recognized that cough, mucus production, airflow obstruction, and shortness of breath on effort were the clinical correlates, in life, of the finding of emphysema at autopsy. He recognized that air trapping and increased collateral ventilation were features of emphysematous lungs and that the peripheral airways were the primary site of obstruction in emphysema. He also speculated that loss of elastic recoil was a factor in diminished flow from the lungs. Interestingly, he recognized that emphysema was associated with chronic bronchitis, which he considered to be the chief causative factor. The relationship between chronic bronchitis and emphysema was also discussed by Louis in 1838,[10] Hasse in 1846,[11] and Waters[12] in 1862. Within a few years, arguments began as to whether the symptoms were always caused by emphysema or whether disease of the conducting airways without emphysema could also cause this syndrome. This is a question that we are still concerned with.

During those same years, the first microscopic descriptions of emphysema appeared, based on plastic casts or on thick sections, which underlined the changes in collagen and elastic fibers. Microscopic examination of the lungs underscored the presence of three consistent findings in emphysematous lungs: the presence of fenestrae in the alveolar walls, the reduction of the capillary bed, and alterations in pulmonary elastic fibers. According to Laennec's theory, hyperinflation of the alveoli appeared to be the first event, and further discussion centered on the mechanisms by which the destruction of the alveolar walls occurred. The predominant opinion was that chronic or repeated hyperinflation led to enlargement of the preexisting interalveolar pores of Kohn or formation of tears in the alveolar wall. Alternative hypotheses favored the role of structural or functional abnormalities of elastic fibers or of compression of the capillaries with resultant ischemic atrophy of the alveolar walls.

Our present knowledge of the pathologic anatomy of pulmonary emphysema dates almost entirely from the middle of the twentieth century, when Gough and Wentworth[13] introduced the use of whole lung sections fixed in inflation and mounted on paper. With this technique, they found "two fundamentally different" types of emphysema, now usually called centrilobular or centriacinar emphysema (CLE) and panlobular or panacinar (PLE) emphysema. Gough's report was based on a study of 140 emphysematous lungs, of which 75 showed CLE.[14] All lungs with CLE had chronic bronchiolitis, and inflammatory changes extended distally to the level where emphysematous expansion occurred. Narrowing of the bronchioles was present in 60% of lesions, whereas dilated or normal airways led to the emphysematous spaces in the remaining 40%. There was no recognizable respiratory bronchiolitis in generalized emphysema (PLE), a finding in marked contrast to that in CLE.

Strikingly absent in all work published prior to 1952 is any illustration of, or reference to, what now might be interpreted as CLE. In view of the excellence of some of the illustrations and the use of inflated lungs by the earlier workers, it seems valid to wonder whether CLE has rapidly increased in frequency in the twentieth century in the same way that bronchogenic cancer has.

The history of chronic bronchitis is far less known than that of emphysema, probably because the term "bronchitis" was used in many different senses. In 1717, Sydenham gave the first description of what could be regarded as chronic bronchitis.[15] Laennec recognized chronic bronchitis, using the term "chronic mucous catarrh," emphasized hypersecretion, and also observed that it could end fatally ("suffocative catarrh"). The term "chronic bronchitis" was introduced by Stokes,[16] who recognized its association with emphysema and also distinguished between mucoid and mucopurulent sputum. Florey and colleagues in 1932[17] described both the increased size of bronchial mucous glands and goblet cell metaplasia as essential features of chronic bronchitis. Reid in 1954[18] provided a comprehensive description of the morphologic alterations of the lungs and airways of patients with chronic bronchitis, with a particular focus on abnormalities of mucous glands, which for many years have been considered a histologic hallmark for the recognition of bronchitis.

The link between cigarette smoking and lung disease took longer to be recognized. Not until the late 1800s did Mendelssohn[19] report that smoking had a deleterious effect on the respiratory system. These early studies were hampered by the lack of sensitive physiologic tests of lung function and relied heavily on differences between smokers and nonsmokers in the prevalence of respiratory symptoms.

Measurements of lung capacity were available after Hutchinson invented the spirometer in 1846. Hutchinson[20] reported a population study in more than 2,000 people of the different subdivisions of vital capacity. Interestingly, Hutchinson believed at the outset that his technique was useful only for the early diagnosis of pulmonary tuberculosis. This indication was taken seriously in many countries, and by 1930 it was still recommended in many European countries to perform spirometry in schools and military compounds for the early detection of tuberculosis.[1]

Not until the 1920s did clinicians become interested in measurements of the disturbance in function that occurs in disease, and the techniques of physiologic measurement moved from laboratory animals to human subjects. Simple spirometers such as Pescher's bottle were commonly used because of simplicity and low cost. The volume of water displaced from the bottle after a forced expiration was a good indication of the vital capacity. With use of these devices, it was then learned that the mean expiratory

volume was decreased in emphysema, pleurisy, fibrothorax, and tuberculosis.[1]

The classic study by Christie in 1933 showed the relationship between lung volume and pressure in normal subjects and that there was loss of lung elasticity in patients with clinical emphysema, a cause of airflow limitation in COPD, as we now know.[21] Shortly afterwards, Cournand and colleagues advanced the notion that emphysema was obstructive.[22] They noted the expiratory slowing that occurred in their patients, as well as the air trapping that occurred as breathing frequency increased. Measurements of the vital capacity ensued, and after years of testing and discussion, the volume fraction of the vital capacity expired in 1 second (the FEV_1) was established, thus forming the basis for the most important present-day physiologic test in the assessment of airflow obstruction. Dayman's[23] classic analysis of expiration indicated that bronchi, bronchioles, or emphysema could obstruct flow.

As clinical pulmonary physiology assumed greater importance and became the dominant science in pulmonary medicine, the previous knowledge of the simple morphology of emphysema was found inadequate, and the usual pathologic methods of examining the lung were deemed unsatisfactory. A better way to visualize and quantitate the disease was needed in order to keep pace with the advances in the understanding of the physiology of the disease. As mentioned above, Gough and Wentworth[13] provided the needed improvement for the study of the disease by developing a technique by which examination of sections of entire inflated lungs became possible and simple. In doing so, Gough[14] laid the foundation of modern knowledge of the pathologic anatomy of pulmonary emphysema. Study of the correlations between the new knowledge of physiologic derangements and the morphology of the disease could now begin.

MORPHOLOGY–FUNCTION CORRELATIONS IN EMPHYSEMA: THE FIRST ATTEMPTS

In the earlier studies correlating morphology and function (1960 to the 1970s), macroscopic diagnosis was used to determine the extent of emphysema, usually in whole lungs obtained at autopsy. Thurlbeck[24] discussed at length the problems associated with the macroscopic diagnosis of emphysema, a subjective method "which may be affected by the opinions and emotions of the observer" and is not improved by the use of point counting. It should also be noted that the standardization of pulmonary function testing had not been achieved in those years. Lung volumes were measured mainly with gas dilution techniques, which may account for some of the differences found among these early studies. However, much work by many authors correlating the morphology and the associated function in emphysema was published between 1960 and the mid-1970s, establishing an overall association between the extent of disease and different functional parameters. The first reported work used maximum voluntary ventilation, a function of the FEV_1, as the physiologic measurement, and it was significantly correlated with the extent of macroscopic emphysema[25] in 31 patients. Two other series found a close relationship, -0.76[26] and -0.74,[27] between the FEV_1% predicted and extent of emphysema, whereas two authors failed to show any significant difference between flows and extent of emphysema.[28,29] Not surprisingly, the best relationships were shown in series that contained high proportions of patients with little airflow obstruction and a wide spectrum of severity of emphysema in their lungs.[26,27,30,31] Correlation coefficients of greater than -0.7 were found in only 3 of the 11 series reviewed here, so it could be concluded that whereas emphysema has an important relationship with expiratory flow rates, this relationship is not precise enough to indicate the amount of emphysema. Subdivisions of lung volumes were no better at predicting the severity of emphysema,[25,26,28–30,32,33] and only the correlation between residual volume (RV)/total lung capacity (TLC) and emphysema was found to be reasonably good by most, but not all, authors.[26,29,33]

The test that all authors have found to be related to the extent of emphysema is the diffusing capacity for carbon monoxide. In the nine series reviewed, correlation coefficients ranged between -0.60 and -0.88. Thus, diffusion capacity remains the test of pulmonary function most closely related to the extent and severity of emphysema. However, as reviewed above, no one test or combination of tests predicts accurately the amount of emphysema in the lungs of living subjects.[34]

We can suitably end this review of the early attempts at correlating lung disease with function in smokers by citing a revealing passage in Thurlbeck[24] heralding things to come. He comments on the remarkable drop in flows found in some patients with trivial emphysema: "We originally attributed this drop to chronic bronchitis, since it seemed unlikely to us that these amounts of emphysema would lower flow rates to this extent. However, the flow rates of other patients with trivial emphysema who had no bronchitis were just as impaired as were those in the patients in the same group who had bronchitis. It thus seems that mild or moderate emphysema may be associated with marked decrease in flow rates."

Not until a few years later, when the concept of small airways dysfunction was re-introduced (Gough had already described the bronchiolitis that accompanied CLE), did an explanation for Thurlbeck's comments start to become apparent.

STRUCTURE–FUNCTION RELATIONSHIP IN NORMAL AIRWAYS: STATIC MEASUREMENTS FOR A DYNAMIC FUNCTION

The small airways of the lung or membranous bronchioles are key in the pathophysiology of COPD. Total airway resistance represents the contribution of the resistance of the various levels of the airway from the larynx and large cartilaginous airways down to respiratory bronchioles. The site of major airway resistance has been studied in dogs[35] and in human lungs[36] with the use of a retrograde catheter technique to partition the contribution of the large cartilaginous and the small airways less than 2 mm in diameter.

These studies revealed that peripheral airway resistance was only a small proportion of the resistance of the entire tracheobronchial tree. In addition, as lung volume changed, total resistance increased near TLC.[37] The low resistance in the small airways was related to their large overall cross-sectional area in the lung periphery. However, other authors, using different techniques to partition central and peripheral resistance, have questioned the original findings[36] and have shown significantly higher peripheral resistance in normal lungs.[37,38]

According to the Poiseuille equation, airway resistance depends upon the number, length, and cross-sectional area of the conducting airways. The number of airways in the normal lung is determined by the pattern of branching, which is established by the sixteenth week of fetal life. The airway length varies considerably from person to person, depending upon age and body size; airway length also varies within an individual, depending upon the phase of ventilation, increasing during inspiration as lung volume increases and decreasing during expiration. Because resistance to airflow in a given airway changes according to the fourth power of the radius of the airway, the cross-sectional area within the tracheobronchial tree is by far the dominant determinant of airway resistance. The cross-sectional area of any given airway is determined by the balance between two opposing forces: those forces tending to distend the walls and those tending to contract the walls, primarily the tension exerted by airway smooth muscle and elastic elements. Airways change in length and diameter during breathing, and in general both dimensions change in proportion to the lung volume. This outward force is provided from the attached lung parenchyma in the case of terminal bronchioles or from the indirect effects of pleural pressure in the case of intrapulmonary conducting airways. Changes in lung volume above functional residual capacity (FRC) have little effect on airway resistance, but between FRC and RV airway resistance increases rapidly and approaches infinity at RV as airway closure occurs. Although it is useful to relate measurements of airway resistance to those of lung volume, the determinant of the relationship between lung volume and airway resistance is the elastic recoil at the given lung volume.[39]

Changes in the geometry of normal bronchi depend not only on the transmural pressure across their walls but also on the distensibility of the elements composing the walls. These constitutive properties of the airway are reflected in the diameter–pressure relationships of the airways. The results of studies fall into two general categories: one in which most of the changes occur at relatively low distending pressures (6 to 10 cm H_2O)[40] and one in which both length and diameter change continuously over the full range of pressures.[41] These discrepant results can be explained by differences in bronchial smooth muscle tone at the time when the experiments were conducted.[42] When bronchomotor tone is absent, the airways become relatively floppy and distend almost fully at low pressures. In contrast, when tone is present, the airways distend more slowly and continuously during inflation.

Airway caliber and, therefore, resistance is also influenced by complex neurohumoral control. Both intra- and extrathoracic airways respond to changes in lung volumes, as described above, but a portion of the lung volume effect is reflexively mediated and is attenuated by prior administration of atropine.[43] In addition, there are active stimuli for both bronchodilatation and bronchoconstriction. Airway caliber is under reflex control through afferent lung receptors and efferent autonomic cholinergic, adrenergic, and "purinergic" nerves. Local changes in gas tension, either hypoxia[44] or hypercapnia,[45] can narrow airways. The resultant decrease in ventilation to areas of low oxygen partial pressure (PO_2) or carbon dioxide partial pressure (PCO_2) represents a compensatory mechanism that serves to promote a match between ventilation and perfusion; as such, it is analogous to, although less important than, hypoxic vasoconstriction.

It is obvious from the above that, no matter how accurate the morphometry and other experimental pathology methods used, the results obtained by analyzing "static" microscopic sections will provide only part of the picture. However, a good morphometric analysis of the possible abnormalities might help reveal the abnormal dynamic behavior of the airways.

MORPHOLOGY–FUNCTION CORRELATION OF THE SMALL AIRWAYS IN SMOKERS

The so-called "small airways" comprise both the membranous bronchioles (less than 2 mm in diameter) and the respiratory bronchioles. When they involve inflammatory processes, the changes are termed "bronchiolitis" and "respiratory bronchiolitis" respectively. The terms "small airways" and "membranous bronchioles," and "small airway disease" and "bronchiolitis," are used interchangeably in this chapter.

As mentioned above, contrary to previous beliefs, Macklem and Mead,[35] by using the novel technique of the retrograde catheter, demonstrated that airways less than 2 mm in diameter (small airways) contributed no more than one-quarter of the total airway resistance in dog lungs. Hogg and colleagues,[36] applying the same technique to human lungs, found that in excised normal lungs only 25% of the total airways resistance was contributed by airways less than 2 to 3 mm in diameter. However, in smokers with mild emphysema, there was a fourfold increase in peripheral airway resistance, whereas total airway resistance remained largely unchanged. More severe degrees of emphysema resulted in a marked increase in total airway resistance due almost entirely to the increase in the peripheral airway component. This work established the then new, and still prevailing, concept that peripheral airways are the major site of increased resistance and disease in smoke-induced obstructive lung disease and that significant increases in the resistance of these airways can be present without measurable changes in the total airway resistance of the lung—hence the term coined by Mead[46] to describe the small airways, "the silent zone of the lung."

Niewoehner and colleagues[47] were the first to demonstrate that definite pathologic abnormalities could already be present in the peripheral airways of young smokers. Membranous bronchioles had denuded epithelium and

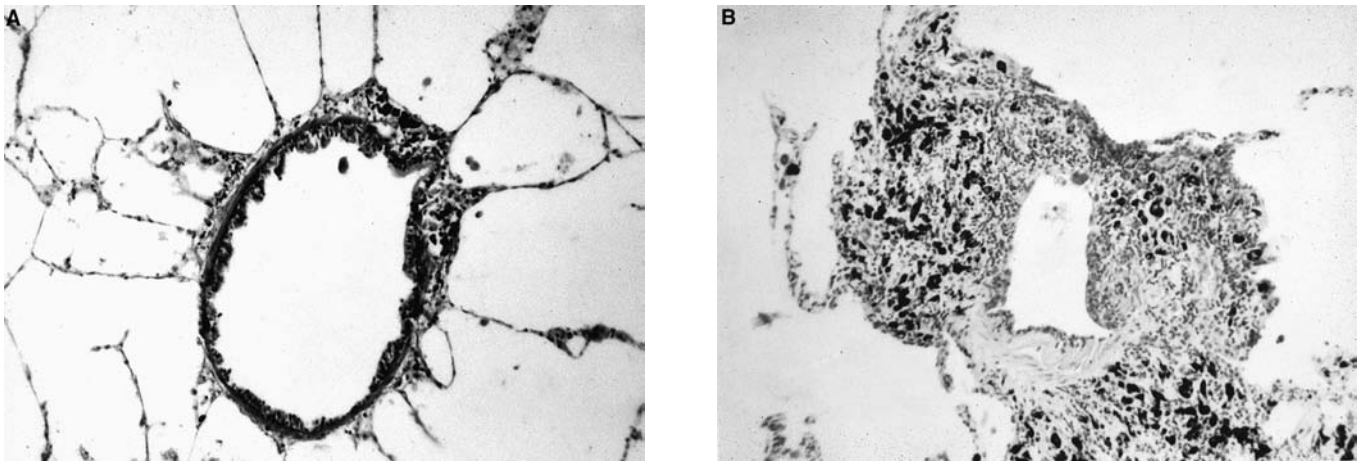

FIGURE 10-1 *A,* Normal small airway. *B*, Abnormal small airway in a smoker (surgical specimen) showing epithelial and wall inflammatory changes. ×100 original magnification.

increased numbers of intramural inflammatory cells. This study demonstrated a definite association between cigarette smoking and pathologic changes in the peripheral airways, and it was hypothesized that these lesions are responsible for the subtle physiologic abnormalities observed in young smokers and may be the precursor of more severe anatomic lesions.

The observations of the early structural changes and site of airflow limitation in smokers in the small airways created enthusiasm for tests of small airway function. The results of such tests, it was hoped, would become abnormal before flow limitation indicated by a decrease in FEV_1 was obvious, and this could identify the smokers most likely to progress to chronic airflow limitation. This was based, in turn, on the hypothesis that structural changes in the small airways preceded the development of COPD. If small airway disease preceded the development of emphysema and COPD, it was not unreasonable to hope that tests of small airway function could be used to identify the susceptible smoker who would develop clinically significant COPD. In order to test this important hypothesis, it was first necessary to demonstrate that the tests of small airway function indeed reflected pathologic abnormalities in the small airways, before performing population studies.

By studying smokers who had tests of pulmonary function, including those sensitive to small airways dysfunction, before undergoing resection for lung tumors, investigators at the Meakins-Christie Laboratories at McGill University[48] developed a pathologic score to describe the microscopic changes in the small airways and to relate them to lung function. Specifically, they scored luminal occlusion, goblet cell metaplasia, squamous cell metaplasia, mucosal ulcers, muscle hypertrophy, inflammatory cell infiltrate, and fibrosis of and pigment deposition in the walls of airways less than 2 mm in diameter. This study showed that these patients had similar but much more extensive abnormalities in the small airways than those described by Niewoehner and colleagues.[47] The abnormalities that could be seen in the older smokers from the McGill study were changes in the epithelium with squamous metaplasia and a chronic inflammatory infiltrate and slight increases in the amount of connective tissue in the walls of the small airways (Figure 10-1). As the pathologic and functional abnormalities progressed, the cellular inflammatory infiltrate changed little, but there was a progressive increase in the amount of connective tissue pigment and muscle in the airway wall. Significant goblet cell metaplasia could be seen in the most severely affected airways (Figure 10-2).

Comparison of the physiologic measurements reflecting small airway abnormalities, such as the nitrogen washout test, volume of isoflow (V_{isoV}) (the volume at which maximum expiratory flows during helium–oxygen breathing and air breathing and ΔV_{max50} become equal), and the results of other function tests, such as the percentage of forced vital capacity in 1 second (FEV_1/FVC), midflow rate, and RV, with the pathologic score showed a progressive deterioration as the score for the morphologic abnormalities increased (Figure 10-3). The striking correlation between the progression of physiologic impairment and the degree of small airway disease suggested that inflammatory changes of the small airways made an important contribution to the functional deterioration seen in COPD, even in the presence

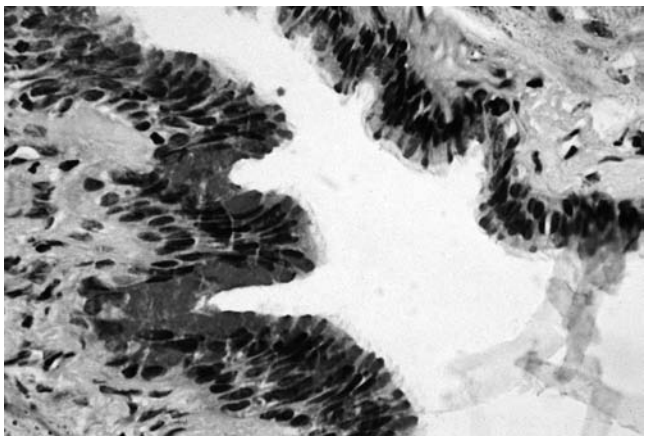

FIGURE 10-2 Goblet cell metaplasia in a small airway of a smoker (surgical specimen). HPS-PAS stain ×250 original magnification.

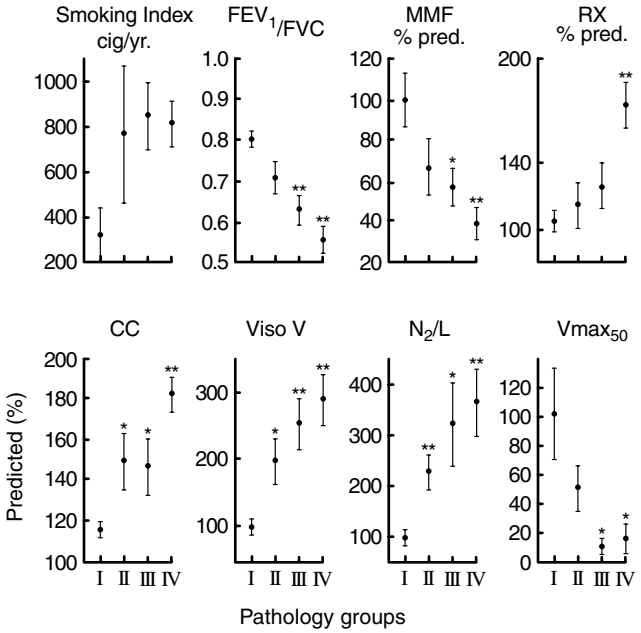

FIGURE 10-3 Correlation of the "small airways" pulmonary functions with the pathologic severity of the small airways (airways score). Cases have been placed into four groups of increasing severity score from I to IV. Reproduced with permission from Hoppin FG et al.[48]

of emphysema. Furthermore, in subjects with normal FEV_1/FVC ratios, two tests of small airway function, the slope of phase III of the nitrogen washout and the volume of isoflow of the air–helium flow–volume curves, were able to detect mild abnormalities of the small airways at a time when the results of other spirometric tests were normal.[48]

Other investigators later confirmed the above findings using lungs obtained either during surgery or at autopsy. Berend and colleagues[49] showed that results of the tests of small airway function correlated best with the total pathologic score of the small airways and, in particular, with the inflammatory score. They also found that the closing volume and the mid–maximum expiratory flow correlated significantly with a measure of airway luminal size.[50] Determining maximal expiratory flow–volume curves in excised human lungs, Berend and colleagues,[51] in another study, showed that flow at the trachea correlated significantly with the total pathologic score of the small airways, as well as with the inflammation, fibrosis, and emphysema scores. Petty and colleagues[52] performed lung function tests on lungs obtained at autopsy and observed that lungs with an abnormal closing capacity had higher total pathologic, inflammation, and fibrosis scores. Wright and colleagues[53] also examined the morphologic basis of small airway obstruction in COPD and attempted to determine the usefulness of tests of lung function in identifying early airway disease. In particular, they divided their group of 96 patients into those with a normal FEV_1 and those with an FEV_1 below 80% predicted. When the FEV_1 was normal, the results of several abnormal tests of small airway function were associated with increasing fibrosis scores in membranous bronchioles and with worsening intraluminal and mural inflammatory scores of respiratory bronchioles (respiratory bronchiolitis).

These studies demonstrated that smokers develop abnormalities of the small airways early in life, that these abnormalities are reflected in vivo by tests sensitive to small airway disease, and that the pathologic abnormalities increase as lung function deteriorates further. The scene was then set to investigate whether abnormalities in the results of tests of small airways early in the lives of smokers could predict those who would eventually develop COPD. Buist and colleagues[54] reported a 9- to 10-year follow-up of two groups of smokers in whom spirometry and the single-breath nitrogen test were used throughout the follow-up period to determine the usefulness of the single-breath nitrogen test in identifying smokers who experienced a rapid decline in FEV_1 and were therefore at risk of developing chronic airflow limitation. Eighty-seven percent of the smokers who developed an abnormal FEV_1 during the follow-up had an abnormal single-breath nitrogen test result and subsequently an increased rate of decline of FEV_1. It was therefore useful in identifying smokers at risk of developing chronic airflow limitation; however, its usefulness was diminished by the high proportion of smokers who had mild functional abnormalities but did not progress to develop chronic airflow limitation.

In summary, cigarette smoke seems to elicit an inflammatory reaction in the membranous and respiratory bronchioles early in life, and this abnormality can be detected by tests of small airway function such as the slope of phase III of the nitrogen washout test. However, the early pathologic and physiologic abnormalities do not progress in all smokers, and physiologic abnormalities in the small airways do not allow us to predict the 15 to 20% of smokers who progress to chronic airflow limitation. Further physiologic deterioration is accompanied by progressive pathologic abnormalities—goblet cell metaplasia, fibrosis, muscle, narrowing of the airways, and emphysema. A complex interaction between airflow limitation and changes in airway structure and in the lung parenchyma, still not well understood today, had emerged.

PROGRESSION OF SMALL AIRWAY ABNORMALITIES IN COPD

The striking correlation between the progression of physiologic impairment and the degree of small airway disease suggests that inflammation of the small airways makes an important contribution to the functional deterioration seen in COPD, even when emphysema is present.

Many studies have since addressed the pathologic changes of the small airways in smokers and their relationship with the flow limitation found in established COPD. Of special interest are studies comparing the airway changes in smokers with various degrees of emphysema and COPD with nonsmokers since they give a perspective not only on the effects of smoking and emphysema but also on the effect of aging on the airways.

In one such study, Cosio and colleagues[55] compared abnormalities in the small airways in smokers and in

nonsmokers who suffered accidental death. Pulmonary function status was not known; however, the degrees of both macroscopic emphysema and microscopic emphysema, assessed with the mean linear intercept, were no different in smokers and nonsmokers, suggesting that, in most cases, the effects of cigarettes were mild. Nonetheless, abnormalities in the membranous and respiratory bronchioles of smokers were quite apparent, with increased numbers of goblet cells and cellular inflammatory infiltrates and muscle and respiratory bronchiolitis, in comparison with nonsmokers. The overall mean diameter of airways less than 2 mm was similar in both groups, but smokers had a significantly greater proportion of bronchioles smaller than 400 μm than nonsmokers, and this proportion was closely related to the total score of airway abnormalities. Several other studies have indicated that there is a better relationship between airflow limitation and the proportion of very narrow airways 0.2 mm and 0.35 mm in diameter.[52,56,57]

Wright and colleagues[58] studied lungs of nonsmokers, smokers, and ex-smokers and reported no significant differences in individual values for total pathologic scores for membranous bronchioles between current and former smokers. Wright and colleagues[59] also found that the wall thicknesses of membranous and respiratory bronchioles for each bronchiolar diameter were increased in almost all size ranges in smokers when compared with lifetime nonsmokers, indicating that smoking is associated with an increase in airway wall thickness independent of airway size and regardless of the presence or absence of emphysema. However, for the same level of function, the degree of airway abnormalities was quite variable, suggesting that other abnormalities, probably elastic recoil losses, influenced the degree of flow limitation.

Hale and colleagues[60] added another group of 18 lungs of patients dying with documented COPD to the population of smokers and nonsmokers in the study of Cosio and colleagues.[55] This study is of interest since it clearly shows the progression of the small airway pathologic changes from nonsmoking older individuals to smokers with mild disease and finally to smokers dying with COPD (Figure 10-4). The cellular inflammatory infiltrate, fibrosis, and muscle in the airway wall increased significantly in a stepwise fashion in the three groups, and, as expected from our initial study, the number of airways less than 400 μm increased accordingly. The mean diameter of the small airways tended to decrease, but the range of diameters was so large that no statistical differences could be found even between patients with the most severe COPD and nonsmokers. A similarly wide range was found for all the airway inflammatory abnormalities measured in the two groups of smokers, indicating that not every smoker reacts in the same way to cigarette smoke and that some smokers are more prone to develop small airway abnormalities than are others.

Not surprisingly, smokers dying of COPD have more emphysema in their lungs than nonsmokers and patients with mild COPD. The degree of emphysema assessed macroscopically correlates with all abnormalities found in the small airways. With this large degree of intercorrelation, it would not be surprising if in severe COPD, the degree of

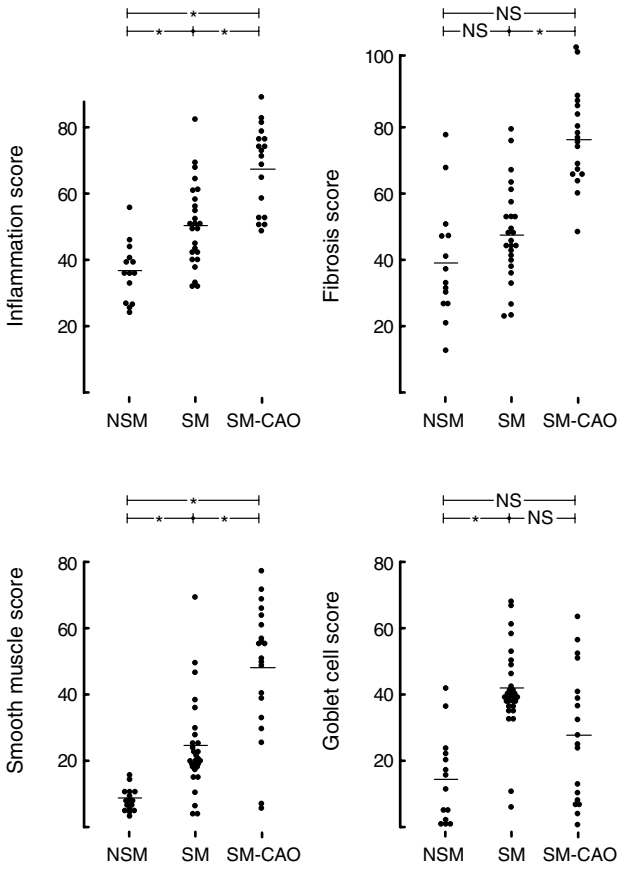

FIGURE 10-4 The progression of the small airway abnormalities in smokers. Groups shown are nonsmokers (NSM), smokers without known chronic obstructive pulmonary disease (COPD) (SM), and smokers with COPD (SM-CAO). All cases are autopsy cases. Reproduced with permission from Cosio MG et al.[55]

emphysema might override correlations between morphology of the small airways and function. Nonetheless, Hale and colleagues[60] found that the degree of airflow limitation correlated not only with emphysema but also with the average airway diameter and the proportion of airways smaller than 400 μm, a function of the total pathologic score of the small airways.

Similar results were obtained by Nagai and colleagues[61] in patients dying with COPD. In their study, flow rates antemortem correlated with the degree of macroscopic emphysema but also with the proportion of airways less than 400 μm in diameter and the degree of deformity of bronchioles. They attributed the decreases in flow to both emphysema causing loss of elastic recoil and airway obstruction caused by an excess number of very small bronchioles and deformity of bronchiolar lumens. It was also clear from this study that, for the same degree of airflow limitation, smokers with lesser degrees of emphysema had more diseased small airways, suggesting that the combined effect of loss of recoil secondary to emphysema and increase in airways resistance secondary to small airway abnormalities produces the airflow limitation in COPD.

The relationship between small airway diameters, a function of the proportion of the very small airways less than 400 μm in diameter in the lung, and lung disease in smokers is of interest. Smokers have a significant excess of

airways less than 400 μm in diameter when compared with nonsmokers, and this proportion correlates with the severity of pathologic abnormalities in these airways, suggesting that the inflammatory reactions in the airways result in an excess of airways less than 400 μm in diameter. However, this explanation may not fully account for the relationship between disease and the caliber of the small airways. The mean bronchiolar diameter and the percentage of airways less than 400 μm in diameter are highly variable even in nonsmokers.[55,62] Small airway caliber may be constitutionally determined or altered because of diseases in childhood, or both, and perhaps persons with smaller airways, because of constitution or childhood illness, are more susceptible to developing disease in these airways when exposed to cigarette smoke or other irritants.

Beyond Morphometry: Mechanisms of Airway Inflammation and Airway Constriction

As discussed above, airway dimensions are smaller in smokers than in nonsmokers. However, these differences seem to be minimal and only evident through calculation of the proportion of airways smaller than 400 μm and, by themselves, may not account for the large increases in airway resistance found in the lungs of smokers. It was explained above how the conducting system of the lung, the airways, does not consist merely of passive tubes, as is assumed for morphometry, but is composed of structures able to change in size during the respiratory cycle and in response to stimuli. It would then seem reasonable to assume that the in vitro measurements of airway diameter might not accurately reflect the airway dimensions in vivo. The main reasons for this discrepancy could be as follows: (1) airway diameters in vitro are measured at TLC, whereas expiratory flows depend on the behavior of the airways during a full lung deflation from TLC to RV, and (2) active constriction of the airways in vivo will not necessarily be evident in fixed human lungs. Hence, the in vitro measurements may not detect these events.

There is ample evidence demonstrating that the airways of smokers react to nonspecific stimuli by constricting, causing an increased resistance and a decreased FEV_1. Many authors[63–69] believe that smokers might have hyperreactive airways and that this hyperreactivity might contribute to the natural history of COPD. Furthermore, the inflammatory abnormalities found in the airways of smokers could contribute to the constriction of normal airway smooth muscle. Hence, regardless of the presence or absence of muscle hyperresponsiveness, muscle constriction in smokers is most likely an important component of airflow limitation in COPD. In support of the role of muscle hyperresponsiveness in the mechanism of COPD are the results of a recent and comprehensive population survey in which smokers showed a relationship between airway responsiveness to methacholine and the rate of decline of lung function[70]; the greater the degree of airway responsiveness at baseline evaluation, the steeper the decline in FEV_1 in response to smoking, a finding consistent with the Dutch hypothesis.

Several pathologic changes could be responsible for active muscle constriction and airway narrowing in COPD. These include (1) airway epithelial damage resulting in increased epithelial permeability and impairment of other epithelial functions, (2) chronic airway inflammation, (3) fibrotic changes in the airway wall, (4) increased amounts of smooth muscle with altered smooth muscle sensitivity and contractility, and (5) loss of alveolar attachments. The role of the epithelium and inflammation may be the key to the pathogenesis of airway narrowing.

Role of the Epithelium in Airway Inflammation and Narrowing

Effects of Altered Permeability The protective barrier formed by the airway epithelium is altered by cigarette smoke. Denuded epithelium, mucosal ulcers, and goblet and squamous cell metaplasia are consistently found in the airways of smokers. Cigarette smoke is cytotoxic to epithelial cells, and the extent of injury is directly related to the concentration of smoke to which the cells are exposed.[71] Numerous studies have shown that cigarette smoke causes airway epithelium to become more permeable to electron-dense tracers, with damage to junctional complexes being demonstrated in smoke-exposed guinea pigs.[72–74] The altered epithelial permeability leaves underlying afferent nerve endings and irritant receptors exposed to bronchoconstrictor and other proinflammatory substances. Thus, the dysfunction of the epithelial cells could contribute to bronchoconstriction and airway inflammation. Finally, altered integrity of the epithelial barrier permits access of plasma exudate to the airway lumen and has mechanical and inflammatory effects on the small airways.

Effects of Luminal Fluid Once across the epithelial barrier, plasma exudate and its associated macromolecules immediately fill the interstices between epithelial projections.[75] Liquid-filled interstices could amplify the degree of luminal compromise in at least two ways. First, the luminal cross-sectional area is reduced as the interstices fill with fluid.[76] Second, plasma proteins may alter the surface tension of the airway lining fluid, and this can further compromise the airway lumen.[77] Macklem and colleagues[78] have reasoned that if the interstices fill with liquid, the surface tension of the airway lining liquid increases, and the radius of curvature of the interfaces joining the tips of epithelial projections increases. At the point where the curvatures of the interfaces joining projections match the curvature of the airway lumen, a point of instability is reached and airways could close. This has been suggested as a physical mechanism that could contribute to airway hyperresponsiveness. Any such effect will be enhanced through impairment of the surface-active properties of surfactant.

Inflammatory Effects of Epithelial Disruption The present evidence suggests that, by sending "danger" signals in response to cigarette smoke, the epithelium is responsible for the initiation and possibly maintenance of the innate inflammatory immune response seen in smokers. Over 2,000 different xenobiotic compounds have been identified

in cigarette smoke, and it has been estimated that there are 10^{14} free radicals in each puff of cigarette smoke,[79] a considerable xenobiotic and oxidant burden on the respiratory epithelium, which is the first line of defense against inhaled substances. Not surprisingly, the defense role comes at a price as cigarette smoke can harm the epithelium and produce apoptosis and necrosis of epithelial cells.[71] There is now ample evidence that, once injured, the epithelium, by increasing permeability and by the production of inflammatory mediators such as interleukin-8 (IL-8), IL-1β, tumor necrosis factor-α (TNF-α), granulocyte–macrophage colony-stimulating factor, and intercellular adhesion molecule-1, could be a potent stimulator of an innate immune reaction.[80,81]

Another consequence of injury to the epithelium caused by cigarette smoke and the resultant increase in epithelial permeability[82,83] is the production and release of tachykinins (substance P and neurokinin-A). These neuropeptides are synthesized by sensory neurons and stored in the terminal parts of the axon collaterals found beneath and within the epithelium, around blood vessels, around submucosal glands, and within the muscle layer of the airways. The release of tachykinins from sensory nerves can be evoked by a variety of stimuli, including cigarette smoke,[84,85] producing bronchoconstriction and modulating a number of important immunologic functions, such as T-cell proliferation,[86] lymphocyte traffic,[87] and production of cytokines, including IL-1, IL-3, IL-6, IL-10, IL-12, and TNF-α.[88,89]

The inflammatory effects of the tachykinins are, in part, controlled by neutral endopeptidase, an enzyme on the surface of the specific cells that are the site of action of tachykinins, and that cleaves and inactivates them. Neutral endopeptidase activity is found specifically in epithelium, glands, nerves, and smooth muscle.[90] The abolition of neutral endopeptidase activity by stripping the epithelium or by inhibition of this enzyme by specific inhibitors is known to increase smooth muscle responses to bronchoconstrictor substances.[91] If airway epithelium is shed or altered, any effects of tachykinins may be more pronounced, not only on airway smooth muscle but also on the inflammatory effects of tachykinins in the mucosa and submucosa.

Endogenous Bronchodilator Effects Respiratory epithelium seems to directly modulate the responsiveness of bronchial smooth muscle by the release of inhibitory factors. In fact, the respiratory epithelium has a high density of β-adrenoreceptors, higher than that in bronchial smooth muscle.[92] The increased smooth muscle relaxation in response to isoproterenol in bronchial rings with intact epithelium compared with preparations denuded of epithelium highlights the importance of the β-adrenoreceptors in the epithelium–smooth muscle interaction.[92,93] Airway mucus, often increased in COPD, may inactivate some epithelium-derived factors.[94]

In summary, the integrity of the epithelial surface seems to be necessary for the normal function and regulation of the airways. Loss of the epithelium and/or possible replacement of normal epithelial ciliated cells by goblet or squamous cells may be one of the initial events in the development of an inflammatory reaction and airway narrowing in smokers. The chronicity of the stimulation could perpetuate the process and enhance the progress of the pathologic changes in smokers.

Airway Wall Contribution to Airway Narrowing

It has been shown that as obstruction worsens, so does the associated degree of airway fibrosis.[48,53,95] This, along with the inflammatory infiltrate, will contribute to the increase in the thickness of the airway wall and the decrease in the thickness of the airway lumen. The amount of muscle in the airway wall is also increased in smokers compared with nonsmokers,[55] and this increase becomes more pronounced as a function of worsening airflow obstruction. It may contribute to airway narrowing through active constriction or through encroachment on the luminal area.

The walls, both internal and external to the smooth muscle layer, are thicker in smokers with airflow limitation than in smokers without airflow limitation, and increasing thickness is associated with smaller airway diameters.[96] Thickening decreases the airway lumen and also has an important effect on the mechanical behavior of the small airways. Moreno and colleagues[97] and Wiggs and colleagues[98] elegantly showed that as the muscle constricts, the thickness of the wall increases, causing the wall to encroach on the lumen in such a way that the thicker the airway wall internal to the muscle, the greater the narrowing resulting from a given degree of smooth muscle shortening. Wiggs and colleagues[98] expanded upon the ideas of Moreno and colleagues and examined the effects of airway wall thickening, loss of elastic recoil, and airway smooth muscle shortening on the increase in airway resistance, using a model of the human tracheobronchial tree. This analysis showed that moderate amounts of airway wall thickening, which has little effect on baseline resistance, can profoundly affect the airway narrowing caused by smooth muscle shortening—especially if the wall thickening is localized to peripheral airways.[96,98]

Fibrosis is not confined to the internal part of the airway but also occurs external to the smooth muscle layer.[97] The mechanical effects of fibrosis and increased airway wall thickness external to airway smooth muscle are different from those of increased thickness of the internal wall. In normal airways, the data of Gunst and colleagues[99] suggest that the lung elastic recoil provides an elastic afterload to peripheral airway smooth muscle and impedes its ability to shorten. This elastic load increases as lung volume increases. According to the interdependence theory, the pressure applied to the outer surface of intrapulmonary airways is equal to the sum of forces applied to the outer surface of the airway by the attached alveolar walls expressed as a fraction of the external surface area of the airway. As the airway wall thickens, the attached alveolar walls become shorter, the forces on the airway wall decrease, the outer airway surface area increases, and hence the pressure applied to the airway decreases.[100] In support of these theories are the findings of Ding and colleagues[101] and Bellofiore and colleagues[102] in elastase-induced emphysema in rats, showing the importance of elastic recoil in smooth muscle shortening in these animals.

Alveolar Attachments

From the previous discussion, it is apparent that loss of the alveolar support around the airways, if it were to occur in smokers, ought to have an important effect on the patency of the small airways and consequently on lung function.

Anderson and Foraker were the first investigators to link pulmonary parenchymal and small airway disease in smokers.[103] They postulated that inflammation of the small airways extends to the parenchyma, leading to weakening of the alveolar walls with destruction. The loss of alveolar attachments to bronchiolar walls will lead to a reduction in the caliber of the airways due to loss of radial traction forces (Figure 10-5). Saetta and colleagues[104] tested this hypothesis and indeed demonstrated a strong association between loss of alveolar attachments and bronchiolar narrowing in the lungs of subjects with mild chronic airflow obstruction. As postulated by Anderson and Foraker,[103] the inflammation of the small airway walls correlated closely with the number of abnormal alveolar attachments.[104] Functionally, the loss of alveolar attachments was found to correlate significantly with loss of elastic recoil, an increase in the closing volume, and a decrease in FEV_1.[104,105] Nagai and colleagues[106] examined lungs obtained at autopsy from patients with moderate-to-severe chronic airflow obstruction and emphysema and found that the loss of normal alveolar attachments was closely related to emphysema. The deformity index, used to describe the irregularity in shape and deformity of peripheral airways, was related to the loss of alveolar attachments and to the decreases in flow in these patients. Thus, it seems apparent that emphysematous changes around the airways will significantly interact with airway patency in the production of airflow limitation.

FIGURE 10-5 Small airway in chronic obstructive pulmonary disease, showing decreased alveolar attachments and narrowed lumen.

STRUCTURE–FUNCTION RELATIONSHIP IN THE LUNG PARENCHYMA

Microscopic Findings in the Lung Parenchyma in COPD: The Doughnut Effect

The traditional method for the study of emphysema has been naked-eye observation (with or without a dissecting microscope or magnifying lenses) of lung slices, either in thin paper-mounted sections or barium-impregnated thick slices floated in water. The focus of these studies has been the identification of holes (or bullae), that is, the terminally destroyed lung in which the type of emphysema producing the hole (CLE versus PLE) is often difficult to identify, especially in severe emphysema cases. There is ample evidence, both functional and microscopic,[107,108] that the so-called normal lung found between the emphysematous holes (the doughnut) is not normal, and microscopic emphysematous lesions have been found. It follows that the "doughnut" will be intimately related to the "hole," that is, the emphysema found under the microscope in the macroscopically apparently normal lung will give rise to the hole as emphysema progresses. Furthermore, in an emphysematous lung, the function will probably be determined in large part by the properties of the apparently normal lung, "the doughnut," found between the holes, something that can be assessed by microscopy. This concept has been elegantly demonstrated by Hogg and colleagues.[109] In excised human lungs with CLE, they measured lung volumes with radiologic methods, in which the emphysematous spaces could be visualized and measured, combined with measurements of pressure, thus obtaining the compliance of the centrilobular emphysematous spaces along with the compliance of the rest of the lung. They found that in the range of RV to TLC, emphysematous spaces changed very little in size with changes in elastic pressure (very low compliance), indicating that there is little if any contribution of these spaces to lung emptying. The mechanical characteristics of the rest of the non–obviously emphysematous lung were much more normal, and the compliance was higher.[109]

It should be evident by now that the pathophysiology of COPD is a complex interplay of pathologic abnormalities in the airways and lung parenchyma. Therefore, in order to understand the deranged function in COPD, we need to study the morphology of the airways and parenchyma and correlate them with the functional abnormalities.

Relationship Between Small Airway Abnormalities and Emphysema in Smokers

Physiologic evidence suggesting that smokers could develop two different types of emphysema was provided by Eidelman and colleagues.[110] They found that smokers with COPD had different patterns of functional abnormalities. Some exhibited pressure–volume curves typical of emphysema, which resembled those seen in α_1-antiprotease deficiency, with high compliance and low elastic recoil pressure at high lung volumes. However, about one-half of their subjects had low or normal compliance and, despite similar elastic recoil, had lower FEV_1 values and pressure–volume curves not typical of emphysema.

Based on these findings, Kim and colleagues[111] reasoned that such dissimilar functional behavior ought to correspond to different parenchymal morphologic abnormalities: PLE for the smokers with mechanical characteristics similar to those seen with α_1-antiprotease deficiency and CLE for the others. They tested this hypothesis in 34 patients undergoing lung resection who had pulmonary function

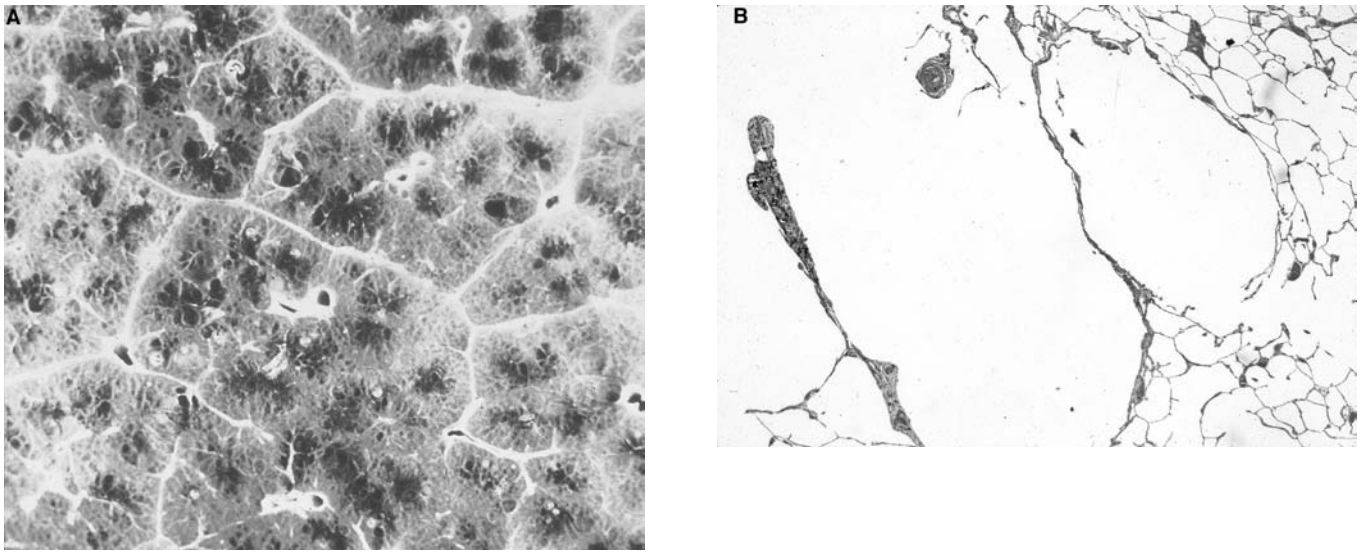

FIGURE 10-6 *A*, Macroscopic view of centrilobular emphysematous areas in the center of the acinus are surrounded by a normal-looking lung. *B*, Micrograph of a lung section showing centrilobular emphysema; similarly, emphysematous destruction is surrounded by normal alveoli.

tests performed before surgery. Emphysema was assessed microscopically and characterized as CLE (Figure 10-6) or PLE (Figure 10-7) with the use of available definitions; it was quantitated by means of the mean linear intercept. Both types of emphysema could be found in this population of smokers; 18 patients had pure or predominant CLE, and 16 had PLE. The mechanical properties were found to be different in both types of emphysema. Patients with PLE had a higher compliance and a higher constant of elasticity (*K*) than patients with CLE. As the emphysema worsened, compliance, *K,* and the elastic recoil at 90% of TLC worsened in PLE but did not change in CLE. Furthermore, losses of elasticity correlated significantly with the extent of emphysema in lungs with PLE but not in those with CLE (Figure 10-8). These findings suggest that mechanical properties are different in the two types of emphysema, and these differences become more marked with the progression of the airspace enlargement, suggesting that the two types of lung destruction are different from the start.

There is further evidence in the literature, beyond the simple microscopic morphology, of the differences between CLE and PLE, and these differences could have important effects on parenchymal function in the two types of emphysema. Substantial evidence exists that elastic fibers undergo proteolytic destruction in the lungs in pulmonary emphysema.[112] Wright[112] compared the three-dimensional architecture of 200 to 300 μm–thick sections of normal and emphysematous lungs and observed that elastic fibers in emphysema were attenuated, separated from each other, and retracted at the unattached ends. Damaged elastic fibers were generally associated with alveolar wall fenestrae, loss of capillaries, and a slight increase in the amount of fibrous tissue. Fukuda and colleagues[113] studied the structure of the elastic network of the lung in CLE and PLE and found important differences between the two emphysema types. In

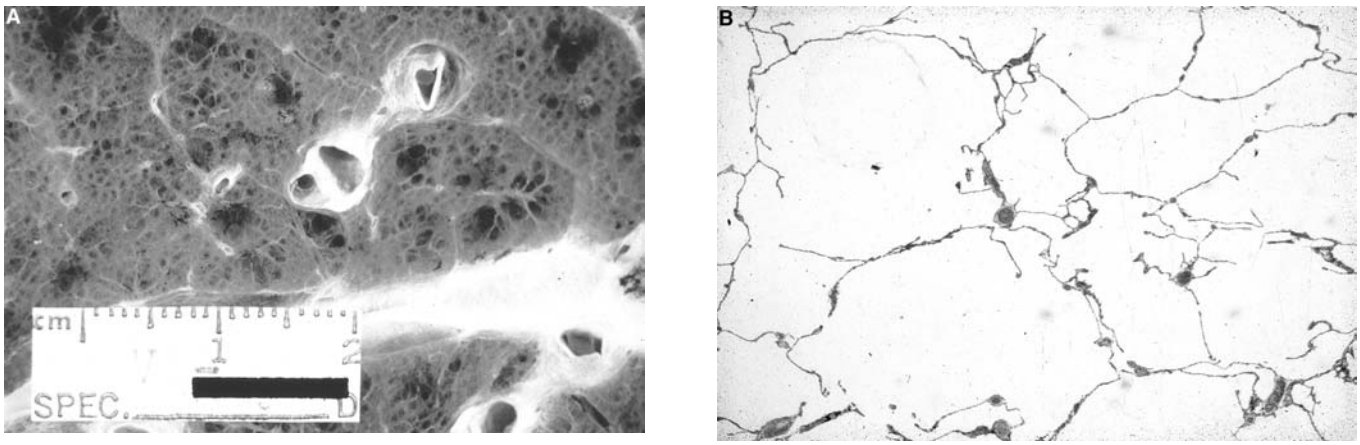

FIGURE 10-7 *A*, Macroscopic view of panlobular emphysema (PLE). The whole acinus is abnormal. *B*, Micrograph of a lung section showing PLE. All airspaces are enlarged, and the size of the alveoli approaches the size of the alveolar ducts.

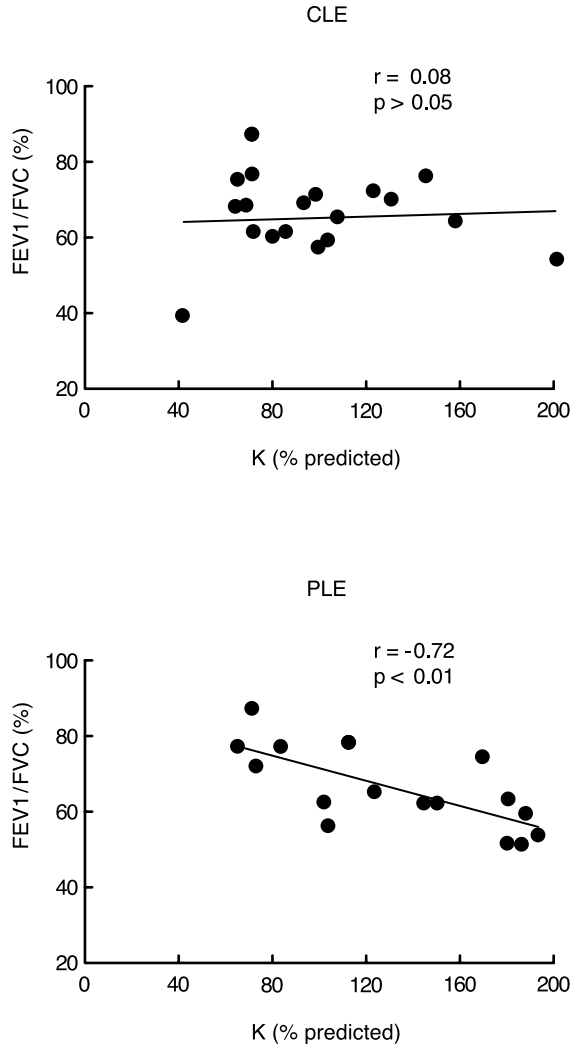

FIGURE 10-8 Correlation between elastic recoil (*K*) and flow (FEV_1/FVC) in smokers with centrilobular emphysema and panlobular emphysema. Pulmonary function was measured prior to surgery in 38 smokers undergoing lung resection for lung nodules. Reproduced with permission from Kim WD et al.[111]

the lungs of patients with PLE, fragmented elastic fibers were observed frequently in alveolar walls and occasionally in bronchioles. At the electron microscopic level, the amorphous component of the elastic fiber appeared to be disrupted, with a fine granularity and a diminished electron density in comparison to normal elastic fibers. In contrast, the lungs of patients with CLE contained few of the disrupted elastic fibers observed in lungs with PLE.

The study of alveolar fenestrae in emphysema also shows how the ultrastructural anatomy of emphysema varies. In our laboratory, we have studied the numbers, size, and distribution of these fenestrae in lungs with CLE and PLE. In CLE, larger fenestrae are found in the center of the acinus, probably destroying respiratory bronchioles and possibly alveolar ducts (Figure 10-9). Fenestrae in the periphery of the acinus are smaller and less numerous. In PLE, however, the fenestrae are smaller than in CLE and distributed equally throughout the acinus.[114,115]

These findings emphasize the distinctiveness of the two types of emphysema that smokers develop. PLE is diffuse,

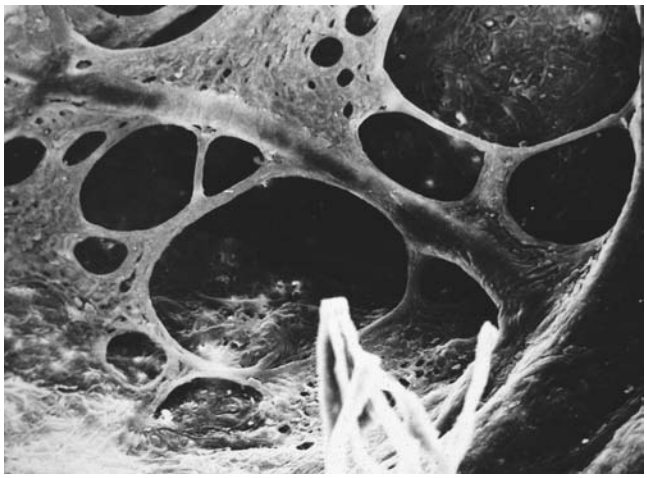

FIGURE 10-9 Scanning electron microscopy of an alveolar wall in a case of centrilobular emphysema, showing prominent fenestrae in the "near" portion of the acinus. Reproduced with permission from Angus GE et al.[114]

affecting all the structures in the acinus evenly and preferentially destroying the elastic network of the lung. In CLE, the destruction is preferentially in the center of the acinus, where inflammatory and mechanical changes (negative pressure generated with lung inflation, more marked in the upper parts of the lung) probably act synergistically to increase the fenestrae size; the fenestrae eventually coalesce, destroying the lung parenchyma. In this type of emphysema, the elastic fibers seem to be less damaged but are rearranged around large emphysematous spaces. Such dissimilar behavior ought to have important functional consequences that are of interest when correlative studies of emphysema and function in smokers are being considered.

Another important difference between the two types of emphysema found by Kim and colleagues[111] was the extent of the small airway abnormalities, later confirmed by Saetta and colleagues[116] and Finkelstein and colleagues.[117] Lungs with CLE had higher total pathologic scores of the small airways than lungs with PLE. The higher scores were mainly ascribable to increased amounts of muscle and fibrosis in the airway wall. Probably as a result of the more severe pathologic involvement, lungs with CLE had more airways smaller than 400 μm in diameter than lungs with PLE (Figure 10-10). Not surprisingly, the pathophysiology of flow limitation in smokers, a function of airway resistance and elastic recoil pressures, differs between the two types of emphysema. Flow decreases as airway abnormalities increase in CLE, but there is no relationship between flow and airway disease in PLE (Figure 10-11). In contrast, flow decreases significantly as elasticity decreases in PLE but not in CLE (Figure 10-12). These findings clarify the pathogenesis of airflow limitation in smokers and indicate that in CLE, loss of flow is primarily a function of airway abnormalities, with elastic recoil loss playing an additive role. By contrast, in patients with PLE, the flow limitation seems to be mainly a function of reduced elastic recoil, and added airway abnormalities could worsen flows even further in these cases.

The studies of Kim and colleagues,[111] Saetta and colleagues,[116] and Finkelstein and colleagues[117] are of interest

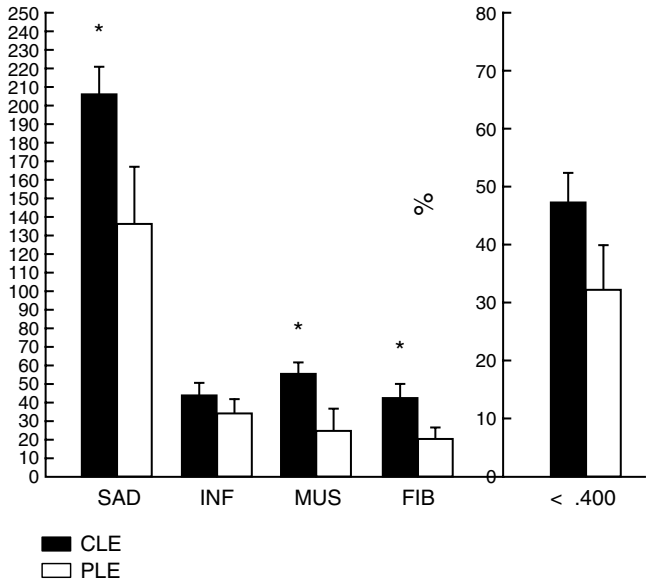

FIGURE 10-10 Pathologic score and proportion of airway diameters smaller than 400 μm in 38 smokers' lungs with either centrilobular emphysema or panlobular emphysema. Reproduced with permission from Kim WD et al.[111]

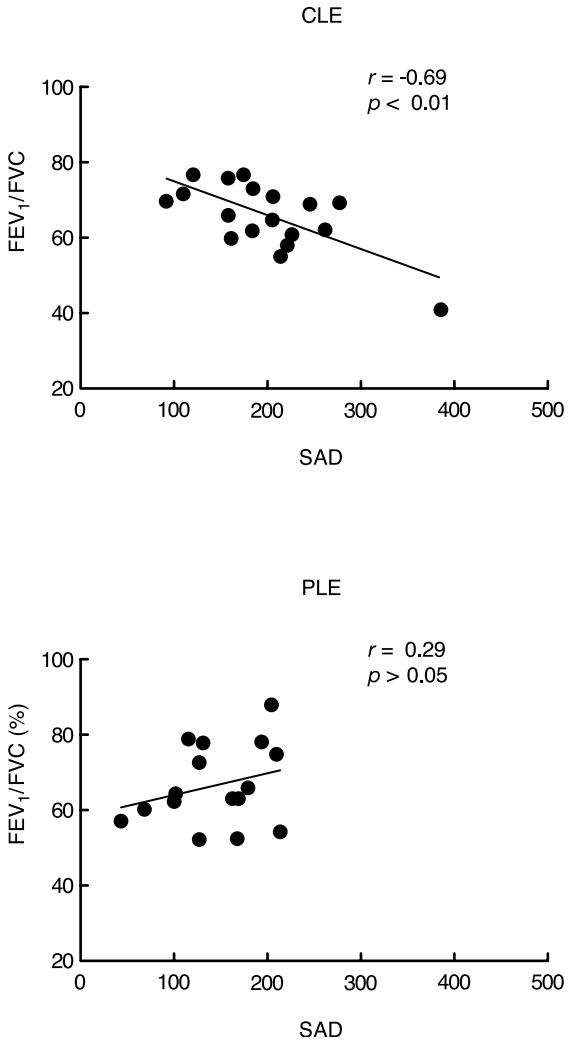

FIGURE 10-11 Correlation between the pathology of the small airways (SAD) and flow (FEV_1/FVC) in smokers with centrilobular emphysema and panlobular emphysema. Reproduced with permission from Kim WD et al.[111]

because they not only clarify the mechanisms of airflow limitation and the role of the bronchioles in COPD but also confirm the possibility of smokers developing two diseases with different pathogenetic mechanisms. The diffuse parenchymal destruction seen in PLE may result from a blood-borne mechanism. On the other hand, the uneven pattern of lung destruction seen in CLE is associated with more severe abnormalities in the small airways, suggesting that centrilobular destruction is related to airborne factors and intimately related to the airway inflammatory process. In support of this hypothesis are the findings of Saetta and colleagues,[116] who found a close correlation between the inflammation of the airways and the parenchymal destruction in lungs with CLE, but not in lungs with PLE. Thus, it is likely that the inflammatory reaction seen in and around small airways and respiratory bronchioles spreads to the parenchyma surrounding these airways and eventually destroys the alveolar walls attached to the airways and the respiratory bronchioles and alveolar ducts. The preservation of alveolar structure and size with concomitant destruction of alveolar ducts and respiratory bronchioles in CLE supports this possibility. Leopold and Gough[118] also showed that the small airways were usually inflamed in lungs with CLE but seldom in patients with predominant PLE. Similar observations were reported by Anderson and Foraker,[119] who felt that CLE and PLE were two different diseases.

These studies provided a new basis for the investigation of lung disease in smokers. If the pathogenetic mechanisms in the two types of emphysema in smokers are different, as the evidence suggests, the study of cigarette-induced lung disease as a single entity will delay even further the understanding of COPD.

Inflammatory Component in COPD: Parenchyma and Airways

The recognition that inflammatory cells play a key role in the pathogenesis of COPD is now so widespread that it has led to the inclusion of the term "abnormal inflammatory response" in the disease definition. In fact, according to the most recent guidelines, COPD is defined as "a disease state characterised by not fully reversible airflow limitation that is usually both progressive and associated with an abnormal inflammatory response of the lungs to noxious particles or gases."[120]

Indeed, the earliest and more constant pathologic abnormality in the airway and parenchyma of smokers is the cellular inflammatory infiltrate. Cellular inflammation, per se, may be responsible for mild airflow limitation,[52,55,94] and it has been suggested that inflammation may lead to functional bronchiolar constriction by releasing bronchoactive mediators.[121] The chronicity of inflammation would, in turn, produce other changes, such as fibrosis of the airway, and could possibly increase the smooth muscle either directly as a result of inflammation or indirectly as a result of chronically increased muscle tone. The stimuli for this abnormality are not precisely known, but it seems that injury to the epithelium, the first structure encountered by cigarette smoke, could promote and perpetuate an inflammatory reaction in the lung.

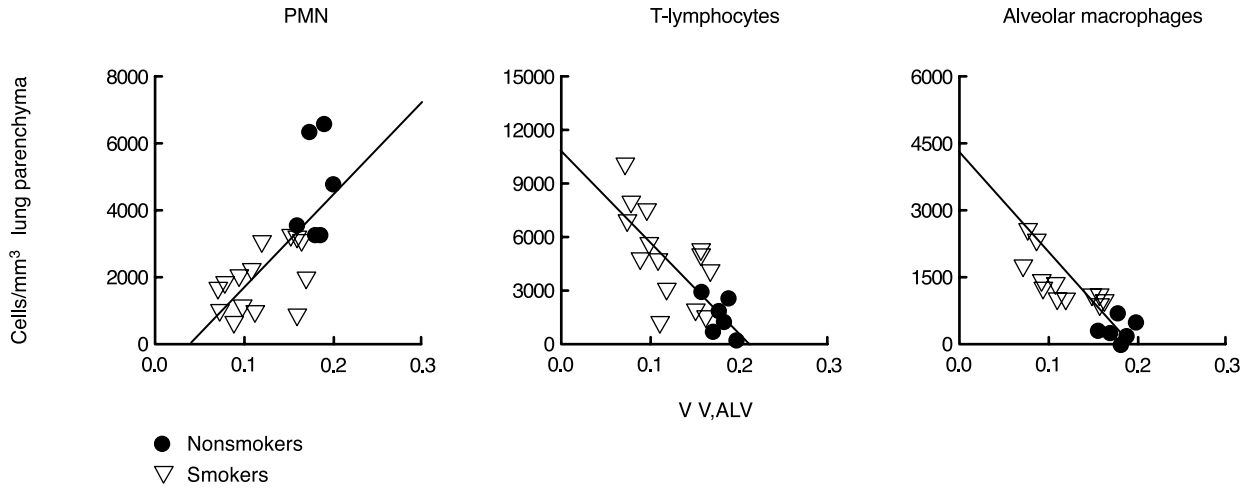

FIGURE 10-12 Correlation of the number of inflammatory cells/mm^3 in the alveolar wall (neutrophils, CD3$^+$ T cells, alveolar macrophages) with the extent of emphysema (V_{VA} = volume of alveolar wall decreases as emphysema increases) in smokers' lungs obtained at surgery. Reproduced with permission from Finkelstein R et al.[117]

In most of the early studies of small airways in smokers, the inflammation of the airways was analyzed in a quantitative fashion. Qualitative morphometric studies of the airways and the lung parenchyma took some time to come, but when they did they changed the way in which we think about the cellular inflammatory mechanism of COPD.

Bosken and colleagues[122] were some of the first investigators to study qualitatively the airway inflammation in smokers with chronic airflow obstruction, and they demonstrated that the number of submucosal neutrophils correlated significantly with the number of cigarettes smoked. They also found that B lymphocytes and CD4$^+$ T cells were most numerous in the adventitia, and there were more CD8$^+$ cells in the epithelium than in the inner wall or the adventitia. B lymphocytes in the entire wall and the adventitia accounted for about 15% of the inflammatory cells and constituted the only cell type that was statistically increased in number in the airways of smokers with obstruction when compared with those without airflow obstruction. Later, Fournier and colleagues[123] and Saetta and colleagues,[124] when studying the large airways of smokers with chronic bronchitis, found increased numbers of CD3$^+$, CD4$^+$, and CD8$^+$ lymphocytes in the bronchial wall. CD8$^+$ lymphocytes predominated over CD4$^+$ lymphocytes, reflecting previously described changes in bronchoalveolar lavage (BAL) fluid and peripheral blood.

Eidelman and colleagues[125] and Saetta and colleagues[126] reasoned that morphometric analysis of the inflammatory cell load in the alveolar wall in smokers would be more informative than BAL fluid. They showed that smokers' lungs had increased cellularity in the alveolar walls, the intensity of which was correlated with the extent of parenchymal destruction. However, the number of neutrophils in the alveolar wall decreased as the alveolar destruction increased, indicating that cells other than the neutrophils were involved in the mechanism of lung destruction. These authors also showed an important correlation between the extent of lung destruction, measured by the destructive index, and losses of lung recoil.

The study that initiated the present interest in the T cell as a possible important cell in the pathogenesis of COPD in general and emphysema in particular was carried out by Finkelstein and colleagues in 1995.[127] Following the findings described above,[125,126] showing a correlation of the number of inflammatory cells, other than neutrophils, with alveolar wall destruction, they investigated which other inflammatory cells could be potentially involved in the production of emphysema. Using immunochemistry, the authors identified the inflammatory cells infiltrating the alveolar wall, and by morphometry they defined the extent of emphysema in smokers and nonsmokers undergoing lung resection. Their findings were surprising at the time as they reported that the most prominent inflammatory cells in the lung parenchyma of smokers were CD3$^+$ T lymphocytes, which increased in number from a mean of 1,546 cells/mm^3 in nonsmokers to 10,000 cells/mm^3 in some smokers. Furthermore, a clear correlation between the number of CD3$^+$ T cells and the extent of emphysema was found, suggesting a role for T lymphocytes in the pathogenesis of emphysema in smokers (see Figure 10-12). Emphysema was also associated with the presence of increased numbers of alveolar macrophages, which significantly correlated with the numbers of T cells, suggesting an interaction between these cells in the inflammatory process leading to emphysema, possibly mediated by a Th1 type of T-cell response (interferon-γ [IFN-γ], TNF-α, and IL-2 cytokine profile). They also confirmed the previous finding that the number of neutrophils in the alveolar wall decreased as the emphysema increased.

Abundant but variable numbers of T cells (CD3$^+$), together with other inflammatory cells, are also found in the small airways of smokers. Finkelstein and colleagues investigated the inflammatory infiltrates in the airways in a series of smokers and nonsmokers in whom airway reactivity had

been measured before lung resection.[117] Smokers were divided according to the type of emphysema in the lung, CLE or PLE. Owing to the large variability in the numbers of inflammatory cells in the airways (from 0 to 500,000/mm^3), no statistical difference was found in the numbers of the different inflammatory cells between the airways of nonsmokers and smokers. However, the degree of airway reactivity correlated with the load of T cells in the airways in smokers with CLE but not in smokers with PLE or nonsmokers. Because similar total numbers of CD3$^+$ T cells were present in the three groups, they suggested that the T cells in CLE were behaving differently, possibly because they were of a different phenotype (Th2 vs Th1). In support of this possibility is the finding that mRNA of the Th2-type cytokines IL-4 and IL-5 (cytokines found in asthma) is abundantly expressed by inflammatory cells in the walls of large airways in smokers with chronic bronchitis and COPD.[128] These cytokines play an important role in the development of asthma, a disease characterized by an increase in airway reactivity; hence, it is possible that clones of T cells in smokers with CLE could express a Th2 cytokine profile that might induce airway reactivity in these cases. Furthermore, IL-4 has been shown to have profibrotic effects by stimulating fibroblasts to proliferate and secrete collagen,[129] and this profibrotic role might be of relevance to the increased remodeling with fibrosis in the small airways and in the increased fibrosis in areas of emphysema found commonly in patients with CLE.[117] Further characterization of the T-cell Th phenotype in the small airways of patients with COPD will be necessary to confirm these possibilities.

Following the report by Finkelstein and colleagues[127] proposing a role for T cells in COPD, Saetta and colleagues identified the CD8$^+$ T cell as the predominant lymphocyte in the small airways[130] of smokers with COPD. This study showed that the only significant difference in the inflammatory cell infiltrate between asymptomatic smokers and smokers with COPD was the increase in CD8$^+$ T cells in patients with COPD. Furthermore, the number of CD8$^+$ T cells was negatively correlated with the degree of airflow obstruction, as measured by the FEV_1, again suggesting a possible role for these cells in the pathogenesis of the disease. This important study introduced the prevailing concept that the development of CD8$^+$ T-cell inflammation in response to smoking is one of the important factors predisposing to the development of COPD. O'Shaughnessy and colleagues found similar T-cell inflammation in the large airways.[131]

Chemokines and T-cell receptors in smokers' lungs have been reported recently by Saetta and colleagues,[132] and their findings indicate that the T cells found in the lungs of patients with COPD are activated, with significant expression of the CXCR3 receptor in the T cells infiltrating the lung, which is not found in nonsmokers. Furthermore, IFN-γ was coexpressed with CXCR3, and the IFN-induced protein-10, a ligand for the CXCR3 receptor, was strongly expressed in the airways and pulmonary arterioles in smokers with COPD but not in other smokers and nonsmoker controls. Importantly, the number of T cells expressing CXCR3, a chemokine receptor restricted to activated T cells and natural killer cells, in the lung[133,134] was correlated inversely with the FEV_1/FVC ratio in smokers, suggesting that as the activated T cells expressing CXCR3 and IFN-γ in the lung increase, there is an increase in lung damage and worsening of the lung function.

The CD4$^+$ T cells are also found, albeit in smaller numbers, in the airways of smokers with COPD, and these cells express activated STAT-4, a transcription factor that is essential for activation and commitment of the Th1 lineage. Not surprisingly, the number of CD4$^+$ T cells expressing activated STAT-4 was associated with the number of cells expressing IFN-γ, and the number of activated STAT-4 lymphocytes correlated with the degree of airflow obstruction.[135] The findings in these two studies strongly support the idea that COPD is mediated by an active Th1 immune reaction in the lung, comprising both CD8$^+$ and CD4$^+$ T cells.

The CD8$^+$ T cells are also the predominant T cells infiltrating the alveolar wall in smokers with COPD, although the numbers of CD4$^+$ T cells are also increased.[136–138] Majo and colleagues,[136] when studying lungs from nonsmokers and smokers obtained at surgery, found that, similar to the airways,[130] the only measurable difference between smokers with and without COPD was a substantial increase in the number of total T cells (CD3$^+$) and CD8$^+$ T cells in the alveolar walls of smokers with COPD (Figure 10-13). Furthermore, the total number of T cells, both CD8$^+$ and CD4$^+$, increased with the number of cigarettes smoked in smokers with COPD but not in healthy smokers, suggesting that the CD4$^+$ T cell is also involved in the inflammatory process in COPD. Majo and colleagues,[136] reasoning that as cytolytic CD8$^+$ T cells were involved, apoptosis should be present, quantitated and found an increased number of apoptotic cells in the lungs of smokers with COPD, which correlated with the numbers of CD8$^+$ cytolytic T cells in the alveolar wall. This and other reports showing an increased number of structural lung cells undergoing apoptosis in emphysematous lungs[139–141] support the idea that CD8$^+$

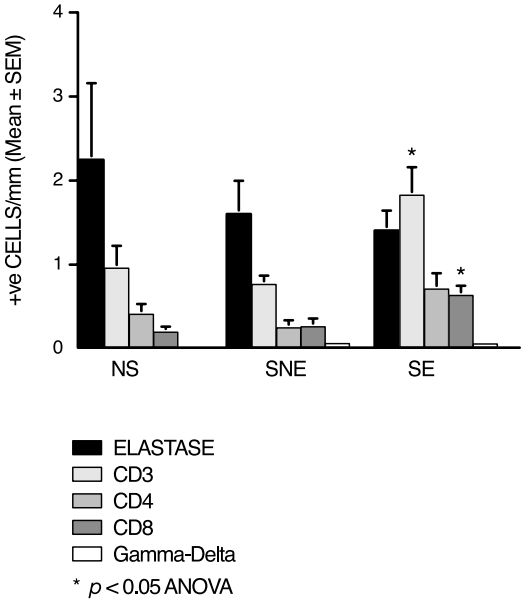

FIGURE 10-13 Cell counts in the alveolar walls of nonsmokers, smokers without emphysema, and smokers with emphysema. All surgical lungs. Reproduced with permission from Majo J et al.[136]

T cells induce apoptosis of endothelial and epithelial cells in emphysema.

Recent studies on smokers with severe COPD demonstrated that the relatively mild infiltration with $CD8^+$ and $CD4^+$ T cells found in patients with mild-to-moderate disease increases markedly in the lungs[138] and airways[142] of severely diseased patients. The numbers of all inflammatory cells, except B lymphocytes, were found to be increased in the lungs of patients with severe emphysema, even though these patients had not smoked more than the control subjects. By far the most numerous cells were $CD4^+$ T cells ($330 \pm 58 \times 10^{12}$) and $CD8^+$ T cells ($250 \pm 51 \times 10^{12}$), but neutrophils, macrophages, and even eosinophils were also increased in number. A relationship between the degree of emphysema and the numbers of each cell type present in the tissue, strongest ($R^2 > 0.8$) for $CD4^+$, $CD8^+$, and alveolar macrophages, was found.[137,138] These studies are of importance because they show that, in COPD, inflammation with an abundance of T lymphocytes and other inflammatory cells continues and even increases late into the disease process.

Recently, we have shown that some mouse strains exposed to cigarette smoke can develop emphysema, and that the severity of emphysema, as assessed by the airspace enlargement and loss of recoil, seems to depend on the ability of the strain to sustain an innate immune response and to mount an adaptive immune response, with $CD4^+$ and $CD8^+$ T cells expressing a Th1 type of cytokine.[143] The finding of animal models that follow the same pattern of disease as humans when exposed to cigarette smoke would be important as it would simplify the investigation of the pathogenic factors in general, and the role of the T cells in particular, in the pathogenesis of COPD in humans.

There is, as we have seen, overwhelming evidence for the presence of T cells in the lungs in COPD and for the activation of these cells. According to the present concepts of T-cell physiology, if the T cells, alone or together with other inflammatory cells, were responsible for the lung injury and progression of COPD, it would be as a response to an antigenic stimulus originating in the lung. Hence, COPD would have to be considered an autoimmune disease triggered by smoking, as previously suggested.[136,144–146]

CONCLUSIONS

Studies correlating the structure of the lung and different measurements of function, from physiologic to cellular and molecular, have been done over the years and are responsible for our present understanding of the pathophysiology of COPD. These studies have revealed that the pathophysiology of COPD is very complex. For the very basic question of how much of the altered function in COPD is due to emphysema, to losses of elastic recoil, and to pathologic and functional abnormalities in the airways, there is not yet a clear answer. However, it is clear that no single one of these abnormalities acts alone and that interactions between the three are responsible.

An important finding is that smokers can develop either CLE or PLE in a pure or clearly predominant form in their lungs in response to cigarette smoking. The different types of emphysema affect the lung parenchyma, the elastic recoil pressure of the lung, and the airways differently. In PLE there are more abnormal and clearly destroyed elastic fibers, more pronounced losses of lung recoil, and less remodeling in the small airways than in CLE. Furthermore, airway reactivity is increased and related to the airway remodeling in CLE but not in PLE. It is our impression that the Dutch hypothesis may well have validity for the patients with COPD who preferentially develop CLE and significant airway remodeling. Thus, COPD should not be considered a homogeneous entity resulting from smoking since CLE and PLE could be fairly different entities, a concept put forward many years ago by Anderson and Foraker.[103] The importance of this paradigm is that if COPD is considered as a single entity, especially from the pathologic point of view, it will take longer to understand the disease. Likewise, studies correlating morphology and function in COPD ought to assess not only the small airways and their abnormalities but also the type and extent of emphysema in order to determine the different contributions of these abnormalities to the pathophysiology of the disease. We believe that such studies would be rewarding.

Looking at the lungs, we have learned to appreciate, quantitate, and identify the cellular inflammatory component in COPD. These studies have broadened the long-standing protease–antiprotease paradigm in the mechanism of emphysema. The discovery of T cells in the lungs of smokers and their correlation with the extent of emphysema and the degree of abnormality in pulmonary function have provided new, more realistic, possibilities for the mechanism of COPD involving an adaptive immune reaction. The consequences of these studies are very important since they are changing the old ideas about the pathogenesis of COPD and are providing avenues for exploration that may explain why only some smokers develop COPD.

We conclude this chapter by inviting readers to consider these last thoughts. If we accept that T cells form part of the inflammatory component of the disease, we have to accept the reason why T cells are in the lung, that is, they are responding to an antigen challenge originating in the lung. If this is the case, we do not think we can escape the conclusion that COPD is a disease produced, at least in part, by antigens (self or modified self) from the lung (autoimmune) secondary to smoking, as we have suggested before.[146] Of course, none of this could be possible without a significant, and persistent, innate immune inflammation, comprising neutrophils and macrophages. Exploring this possibility may lead us to a better understanding and thus to new, and perhaps more effective therapeutic approaches to the disease.

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