

REVIEW ARTICLE

Airway Hyperresponsiveness as a Determinant of the Early Asthmatic Response to Inhaled Allergen

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HISTORY

Atopy and Asthma

Seasonal allergic rhinitis and asthma initially referred to (incorrectly) as *rose catarrh* was ill understood and still the subject of case reports less than 200 years ago (1). In his famous treatise in 1873, Charles Blackley clearly identified pollen as at least one cause of allergic airway symptoms (2). Subsequent studies led to the development of allergen skin testing (3), allergen injection therapy (4), passive transfer of sensitization by serum (5), and the eventual discovery that the serum factor previously termed *reagin* was a newly recognized immunoglobulin called immunoglobulin E (IgE) (6). In subjects who appear equally allergic, allergen exposure may induce allergic rhinitis (nose and eye symptoms primarily), allergic rhinitis and asthma, or infrequently asthma alone. These discrepancies led to the concept that there must be a difference or a susceptibility in the so-called *shock organ* that led to some subjects developing asthma and others rhinitis (7).

Airway Hyperresponsiveness

About 60 years ago, Robert Tiffeneau used new technology that added a time axis to spirometric measurements of lung volumes and measure (expiratory) *flow rates*. Tiffeneau described what we now call the FEV₁ (8). He also used this new technology to measure airway responsiveness to bronchodilators (isoproterenol) and bronchoconstrictors (acetylcholine) and postulated that these could be useful measurements in the clinical evaluation of lung disease (9). By the mid- to late 1950s, Tiffeneau had described, in the French literature, a major understanding of the features of airway responsiveness and indeed described a number of things that have taken a number of decades to rediscover. In 1958, Tiffeneau stated that the degree of allergic sensitivity of the lungs (synonymous with asthmatic response to inhaled allergen) was dependent on two independent factors, first the degree of pulmonary allergy and, second, the degree of airway hyperresponsiveness (10). In 1967, Zuidema stated “if the bronchial

sensitivity to histamine and the result of skin test with allergen to be tested are known, it should be possible to predict with a reasonable degree of certainty the result of the allergen inhalation test” (11). These authors suggest that the difference in the *shock organ*, namely the lung, in allergic asthmatics is the degree of airway hyperresponsiveness. Zuidema did present data suggesting subjects with greater airway hyperresponsiveness responded to lower concentrations of allergen (11).

The current series of investigations were carried out to look at the relative roles of airway responsiveness to histamine (or methacholine) and the degree of allergic sensitization as determinants of the early asthmatic response (EAR) to inhaled allergen.

CLINICAL STUDIES

Our first study investigating this relationship was carried out in 1975 and published in 1976 (12). We studied nine subjects who had allergen challenges in the laboratory. Non-allergic airway responsiveness was performed using the standardized 2-min tidal breathing histamine inhalation test (13). At this time, the concept of expressing airway responsiveness in a continuous fashion as the provocation concentration or dose producing a 20% FEV₁ decrease (PC₂₀, PD₂₀) had not yet been established. We expressed airway responsiveness as an *index* by dividing the percent decline in FEV₁ by the concentration of histamine. This index is, in fact, analogous to dose-response slope, which is now often examined in epidemiologic studies (14). Values of the index increase with increasing airway responsiveness (compared to PC₂₀, which decreases as airway responsiveness increases). We examined the skin test produced by the same concentration of the same allergen extract that caused a 20% FEV₁ early asthmatic response (EAR). We reasoned that if this concentration of allergen produced a large skin test wheal, then it would produce a similarly large allergic reaction with a proportionately large release of mediators in the airway. Thus, we used the skin prick wheal size of the allergen concentration which caused a 20% EAR as an index of the magnitude of the airway allergic reaction. Linear regression of the log airway responsiveness index with the magnitude of the allergic reaction (skin test wheal size) revealed a strong inverse correlation ($r = -0.96$, $p < 0.001$) (12). This indicated that in subjects with mild airway responsiveness, a large airways

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allergic reaction was required to induce an EAR. By contrast, in those subjects with more severe airway responsiveness, a comparatively small allergic reaction was required in the airways in order to induce a significant EAR. Interestingly, the late asthmatic response (LAR) showed no such relationships. There were a couple of problems that hindered the dissemination of these results. First, airway responsiveness was expressed in a way that was not standard. Second, we found that the concept of utilizing the skin test wheal size of the same allergen as an index of the severity of the airway allergic reaction was difficult for colleagues to understand.

To address some of these issues, we performed a second study in 25 subjects in 1979 (15). Airway responsiveness to histamine was measured by the 2-min tidal breathing method and results expressed as the histamine PC₂₀ (13). We examined the degree of allergy to the allergen used for inhalation by the skin test endpoint defined as the smallest concentration of allergen producing a 2-mm wheal. The skin test endpoint is known to correlate well with the level of allergen specific IgE in the serum (16, 17). We also examined the magnitude of the airway allergic reaction as in our first study by using the mean wheal diameter of the concentration of allergen, which caused a 20% FEV₁ decrease when inhaled. The results showed a very strong correlation (Figure 1, $r = 0.91$) between the magnitude of the airway allergic reaction and the log of the histamine PC₂₀, thus confirming our earlier study, which used a less conventional continuous measure of airway responsiveness. We also performed single and multiple linear regression analyses with the allergen PC₂₀, the skin test endpoint and the histamine PC₂₀ (all values logarithmically transformed). We found a strong correlation between the allergen PC₂₀ and the skin test endpoint ($r = 0.60$, $p < 0.005$) and between the allergen PC₂₀ and the histamine PC₂₀ ($r = 0.52$, $p < 0.01$). When the (log) allergen PC₂₀ was expressed as a function of the (log) skin test endpoint and the (log) histamine PC₂₀ with multiple linear regression, the sig-

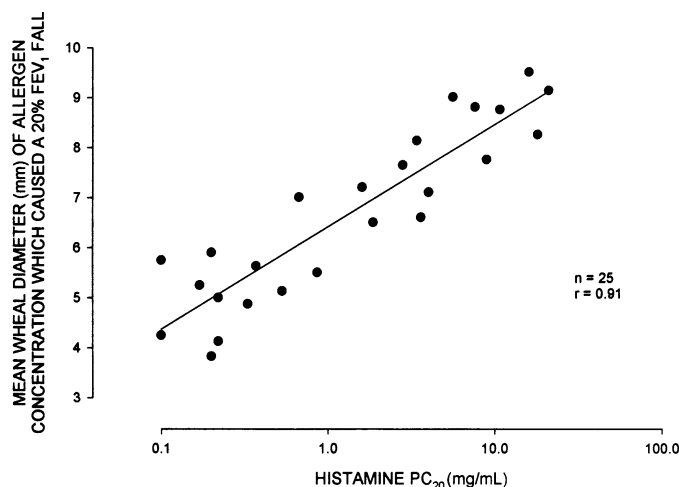


FIGURE 1.—Histamine PC₂₀ (mg/mL) log scale on horizontal axis graphed against the skin mean wheal diameter (mm) of the concentration of allergen, which caused a 20% FEV₁ decrease EAR when inhaled on the vertical axis. Modified from and reprinted with permission (15).

nificance was improved ($r = 0.78$, $p < 0.001$). These results confirmed that the EAR to inhaled allergen was dependent on both the nonallergic airway responsiveness as measured by histamine PC₂₀ and the degree of allergen sensitization as measured by the skin test endpoint. The multiple linear regression equation also provided a means whereby we could predict the allergen PC₂₀ from skin testing and histamine challenge.

Our third study in 1987 was performed to evaluate the performance of this predictive equation (18). We studied 26 new subjects and predicted the early asthmatic response (allergen PC₂₀) from the equation. We found that the allergen PC₂₀ could be predicted to the nearest doubling concentration in 6, and was out by 1 doubling concentration in 11, by 2 doubling concentrations in 6, by 3 doubling concentrations in 3, and by more than 3 concentrations in none. From the total of 50 subjects (one subject who participated in both studies was not counted twice), a new regression was developed. Since the regression coefficients were similar when we did single linear regression of the allergen PC₂₀ against the histamine PC₂₀ and against the allergen skin test endpoint, we elected to express the allergen PC₂₀ in single linear regression against the product of the histamine PC₂₀ and the allergen skin test endpoint. This gave us a much easier regression line ($y = 0.68x + 0$). We published this as a new and simpler predictive equation: $\log \text{allergen PC}_{20} = 0.68 \log (\text{histamine PC}_{20} \times \text{skin test endpoint})$. These data are shown in Figure 2. There are two important caveats for the use of this formula. First, the allergen challenge and the histamine challenge must be done using the identical methods; i.e., same nebulizer, same output, same method of inhalation; it does not really matter which method. Second, the allergen extract and the dilutions thereof used for inhalation must be identical to those used for the skin testing.

For the past almost 20 years, we have been successfully using this formula to predict the allergen PC₂₀ primarily

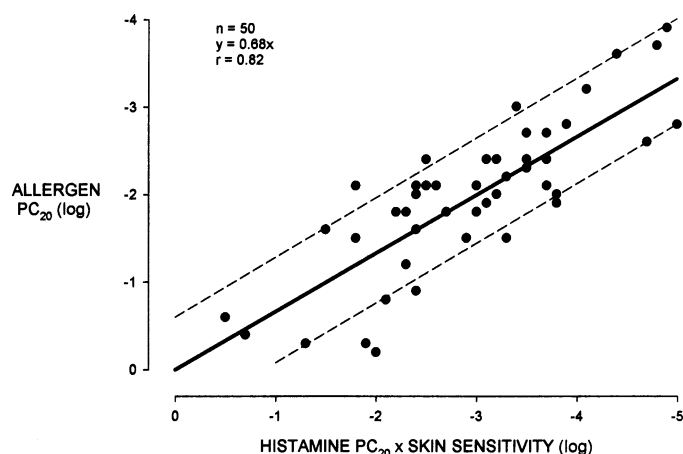


FIGURE 2.—Log (histamine PC₂₀ × skin test endpoint) on the horizontal axis graphed against log allergen PC₂₀ on the vertical axis. Allergen PC₂₀ is expressed as log₁₀ (allergen dilution from stock solution), e.g., 0 = undiluted, -1 = 1:10, ... -4 = 1:10000, etc. Histamine PC₂₀ is expressed in mg/mL and skin test endpoint as allergen dilution analogous to the allergen PC₂₀. The solid line is the regression line; the dashed lines are + and -2 doubling dilutions. Modified from and reprinted with permission (18).

to help establish a starting concentration for allergen challenges in research investigations. However, we have been using the methacholine PC₂₀ rather than the histamine PC₂₀. Histamine and methacholine tend to produce similar responses at the same dose (19); however, the relationships noted above had not been objectively confirmed using the methacholine responsiveness. Because of this, we recently completed a retrospective survey in 158 allergen challenges performed at three centers in Canada who used the same methodology. The results were similar to those obtained with histamine. The log allergen PC₂₀ correlated with log methacholine PC₂₀ ($p = 0.0015$) and the skin test endpoint ($p < 0.00005$). Multiple regression was highly significant at $r = 0.61$ ($p < 0.00005$). Our simple histamine-based formula predicted the allergen PC₂₀ to within two doubling concentrations in 80% of challenges and to within three concentrations in 92% of challenges; this improved slightly by the multiple linear regression methacholine-based formula to 81 and 94%, respectively (20).

DISCUSSION

This series of investigations clearly confirms that the allergen-induced EAR is dependent on the degree of allergy (i.e., the level of allergen-specific IgE), and the degree of nonallergic airway hyperresponsiveness. Other investigators have also demonstrated this. Bryant and Burns documented that serum allergen-specific IgE and airway responsiveness to histamine were both correlated with the airway responsiveness to inhaled house dust mite allergen (21). Hill et al. documented that airway response to allergen could be predicted by airway response to exercise and skin test endpoint to allergen (22). Other investigators have found variable relationships between airway response to allergen and degrees of allergen or degrees of airway responsiveness (23). This likely depends on subject selection. If subjects have a relatively narrow range of airway responsiveness to histamine or methacholine and a wide range of allergen-specific IgE, then it is likely that the relationship between airway response to allergen will seem to be mainly dependent on the level of allergen specific IgE. Indeed, we see this in our 158 subjects studied recently where the airway response to methacholine was in a relatively narrow range compared to airway response to allergen and skin response to allergen and indeed the airway response to allergen correlated better with the skin test endpoint than with the methacholine PC₂₀. By contrast, when subjects with similar cutaneous sensitivity and a relatively wide range of airway responsiveness to histamine or methacholine are studied, the latter may seem to be more important; this may be the explanation for results of some other investigators.

CLINICAL RELEVANCE

These investigations by ourselves and others have clinical relevance in three areas. First, as noted above, airway responsiveness to nonallergic stimuli (histamine, methacholine, exercise) is an important determinant, along with the degree of allergen sensitivity, of the airway response to allergen. Since it has recently been demonstrated that airway response to indirect challenges may correlate better with asthma activity, severity, inflammation, etc. (24–26), then this relationship

might be even better if we used indirect challenges such as adenosine monophosphate (AMP) or mannitol.

Second, if it is clear that the airway response to allergen can be predicted by relatively simple measurements such as the histamine/methacholine PC₂₀ and the skin test to allergen, then the previously held concept that allergen challenges might be useful *to determine the clinical relevance of allergens* (27) would seem not to be necessary. In fact, we strongly believe that allergen challenges in the laboratory should be limited to research investigations either into the mechanisms of asthma or in the investigation of pharmacologic agents. It is worth noting that neither bronchoprovocation with allergen nor accurate prediction of the airway response to allergen can overcome uncertainties related to less than optimal allergen standardization.

Third, we have found the ability to predict the allergen PC₂₀ to within two or three doubling concentrations valuable in helping to safely select a starting concentration for our research allergen challenges in the laboratory. This prevents some overly long allergen challenge.

SUMMARY

In summary, we have done a series of investigations over the last 30 years documenting that the airway response to allergen is dependent on both the level of the allergen specific IgE and the degree of underlying airway hyperresponsiveness as measured by histamine or methacholine challenge. This validates the hypotheses predicted by Robert Tiffeneau who must be regarded as *the father of airway responsiveness*. These data indicate that there is little, if any, role for allergen challenges in determining the clinical relevance of allergens. These data are also useful in helping to safely shorten research allergen challenges.

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REFERENCES

1. Bostock J. Case of a periodical affection of the eyes and chest. *Medico Chirurg Trans* 1819; 10:161–164.
2. Blackley CH. In: *Experimental Researches on the Cause and Nature of Catarrhus Aestivus (Hay-Fever or Hay-Asthma)*; Balliere Tindall & Cox: London, 1873.
3. Casoni T. La diagnosi biologica dell'echinocosi umana mediante l'intradermoreazione. *Folia Clin Chimica Microscopia* 1911; 4:5.
4. Noon L. Prophylactic inoculation against hay fever. *Lancet* 1911; 1:1572.
5. Prausnitz C, Kustner H. Studien uber die Ueberempfindlichkeit. *Centralblatt fur Bakteriologie, Parasitenkunde und Infektionskrankheiten* 1921; 86:160–169.
6. Ishizaka K, Ishizaka T. Identification of gamma-E-antibodies as a carrier of reaginic activity. *J Immunol* 1967; 99:1187–1198.
7. Feinberg SM, Stier RA, Grater WC. A suggested quantitative evaluation of the degree of sensitivity of patients with ragweed pollinosis. *J Allergy* 1952; 23:387–394.
8. Tiffeneau R, Pinelli A. Regulation bronchique de la ventilation pulmonaire. *J Fran Med Chirug Thorac* 1948; 2: 221–244.
9. Tiffeneau R, Beauvallet M. Epreuve de bronchoconstriction et de bronchodilatation par aerosols. *Bull Acad Med* 1945; 129:165–168.
10. Tiffeneau R. Hypersensibilite cholinergo-histaminique pulmonaire de l'asthmatique. *Acta Allerg* 1958; (suppl) 5:187–221.

11. Zuidema P. Value of inhalation tests in bronchial asthma. 2nd International Symposium-Tuberculosis, Climate, Asthma, Chronic Bronchitis, Davos 1967. *Respiration* 1969; (suppl) 26:141–150.
12. Killian D, Cockcroft DW, Hargreave FE, Dolovich J. Factors in allergen-induced asthma: Relevance of the intensity of the airways allergic reaction and non-specific bronchial reactivity. *Clin Allergy* 1976; 6:219–225.
13. Cockcroft DW, Killian DN, Mellon JJA, Hargreave FE. Bronchial reactivity to inhaled histamine: A method and clinical survey. *Clin. Allergy* 1977; 7:235–243.
14. O'Connor G, Sparrow D, Taylor D, Segal M, Weiss S. Analysis of dose-response curves to methacholine: An approach suitable for population studies. *Am Rev Respir Dis* 1987; 136:1412–1417.
15. Cockcroft DW, Ruffin RE, Frith PA, Cartier A, Juniper EF, Dolovich J, Hargreave FE. Determinants of allergen-induced asthma: Dose of allergen, circulating IgE antibody concentration and bronchial responsiveness to inhaled histamine. *Am Rev Respir Dis* 1979; 120:1053–1058.
16. Bryant DH, Burns MW, Lazarus L. The correlation between skin tests, bronchial provocation tests and the serum level of IgE specific for common allergens in patients with asthma. *Clin Allergy* 1975; 5:145–157.
17. Stenius B, Wide L, Seymour WM, Holford-Strevens V, Pepys J. Clinical significance of specific IgE to common allergens. I. Relationship of specific IgE against *Dermatophagoides* spp. and grass pollen to skin and nasal tests and history. *Clin Allergy* 1971; 1:37–55.
18. Cockcroft DW, Murdock KY, Kirby J, Hargreave FE. Prediction of airway responsiveness to allergen from skin sensitivity to allergen and airway responsiveness to histamine. *Am Rev Respir Dis* 1987; 135:264–267.
19. Juniper EF, Frith PA, Hargreave FE. Airways responsiveness to histamine and methacholine: Relationship to minimum treatment to control symptoms of asthma. *Thorax* 1981; 36:575–579.
20. Cockcroft DW, Davis BE, Boulet LP, Deschesnes F, Gauvreau GM, O'Byrne PM, Watson RM. The links between allergen skin test sensitivity, airway responsiveness and airway response to allergen. *Allergy* 2005; 60:56–59.
21. Bryant DH, Burns MW. Bronchial histamine reactivity: Its relationship to the reactivity of the bronchi to allergens. *Clin. Allergy* 1976; 6:523–532.
22. Hill DJ, Shelton MJ, Hosking CS. Predicting the results of allergen bronchial challenge by simple clinical methods. *Clin Allergy* 1982; 12:295–301.
23. Popa VT, Garfield M. Immunologic and pharmacologic determinants of bronchial responses to inhaled ragweed. *J. Allergy Clin. Immunol.* 1979; 63:141–142.
24. Pauwels R, Joos G, Van Der Straeten M. Bronchial hyperresponsiveness is not bronchial hyperresponsiveness is not bronchial asthma. *Clin. Allergy* 1988; 18:317–321.
25. Van Schoor J, Joos GF, Pauwels RA. Indirect bronchial hyperresponsiveness in asthma: Mechanisms, pharmacology and implications for clinical research. *Eur Respir J* 2000; 16:514–533.
26. Joos GF, O'Connor B, Anderson SD, Chung F, Cockcroft DW, Dahlen B, DiMaria G, Foresi A, Hargreave FE, Holgate ST, Inman M, Lotvall J, Magnusson H, Polosa R, Postma DS, Riedler J. ERS Task Force. Indirect airway challenges. *Eur Respir J* 2003; 21:1050–1068.
27. Spector S, Farr R. Bronchial inhalation challenge with antigens. *J. Allergy Clin. Immunol.* 1979; 64:580–586.