

Immunoprophylaxis of atopy: light at the end of the tunnel?

Patrick G. Holt

Current research in the field of atopy is directed almost exclusively towards treatment of established allergic disease. In particular, treatment has concentrated on controlling the release and actions of various mediators, such as cytokines, from the allergy effector cells at the end of the immunoinflammatory cascade. Here, Patrick Holt argues that a potentially more-effective and achievable goal may be the prevention of initial T helper 2 (Th2)-cell sensitization to environmental allergens during infancy. This might be achieved via amplification of the endogenous 'immune deviation' mechanism(s) that normally facilitate discrimination between pathogenic and non-pathogenic antigens at the mucosal surfaces of the body.

Atopy is the principal etiological factor in asthma¹. For reasons that are still unclear, the prevalence of asthma is in the process of an unprecedented upswing in many of the developed countries². Of greater cause for concern, the disease also appears to be increasing in severity². The economic impact of atopy is staggering. During 1990, treatment costs for asthma alone accounted for \$6.4 billion US dollars of the US health bill³, and in the order of one billion pounds sterling in the UK (Ref. 4); and atopic dermatitis and allergic rhinitis add substantially to this burden.

Current treatment regimes emphasize pharmacological control of the inflammatory processes that are now recognized as the major pathogenic factors in atopy. These are largely targeted at the end-stage effector mechanisms responsible for tissue damage in chronic disease. Indeed, reliable and safe immunotherapy for control of the underlying immune responses, which are the initial triggers of the inflammatory 'cascade' in atopy, remains a yet-to-be-achieved goal. Nevertheless, the rapid pace of development in the key areas of cytokine regulation and molecular cloning of allergens gives cause for cautious optimism for the future.

The potential benefits of effective treatment strategies targeted at the upstream immunological events in diseases such as asthma, and the consequent release of sufferers from long-term reliance on anti-inflammatory drugs, are self-evident. The general acceptance of this premise amongst experimental immunologists provides the impetus and the direction for current research in this area. However, this concentration of immunological research resources into areas relating solely to the pathogenesis of established atopy assumes that the more-desirable long-term goal, i.e. prevention of the initial onset of atopic disease, is not practically achievable by employing current immunological tools. This view is challenged below. Here, it is argued that evolution has already shaped a highly effective mechanism for selective downregulation of potentially pathological aspects of the immune response to ubiquitous en-

vironmental allergens. This mechanism forms the basis for long-term 'protection' of the majority of individuals against allergic sensitization to ubiquitous food and inhalant allergens, and the occasional failure of this process is responsible for the inappropriate T-cell responses that lead to IgE production against these antigens in atopic individuals. It is further argued that the general mechanism(s) by which this process operates is sufficiently understood for us to contemplate active *in vivo* intervention in order to prevent failure in individuals who are genetically at high risk of developing atopic disease. This conclusion derives from the recent literature on the etiology of human atopy: in particular the seroepidemiology of IgE responses in normal and atopic children, as compared with studies on *in vivo* and *in vitro* T-cell reactivity to environmental allergens, both in humans and experimental animals.

T-cell regulation of IgE production

Current concepts of T helper (Th)-cell regulation of IgE synthesis derive from studies in the murine system, which have demonstrated that the magnitude and duration of IgE responses are largely determined by the interplay between antagonistic interleukin 4 (IL-4) and interferon γ (IFN- γ) signals⁵. The balance between these cytokines is a key determinant in the *in vivo* expression of the IgE-responder phenotype^{5,6}.

In the mouse, these cytokines appear to be produced by distinct Th-cell subsets derived from common uncommitted Th0 precursors, which have been designated Th1 and Th2 (Ref. 7), and it is apparent that a similarly biased distribution of cytokine profiles exists amongst Th-cell subsets in humans⁸⁻¹⁰. Furthermore, these cells regulate IgE production by human B cells *in vitro* in a fashion that is comparable with that reported for Th cells in murine systems^{11,12}.

It has subsequently become evident from *in vitro* studies in human and animal systems that a variety of cytokines from other cell populations also play important roles in this process. IFN- α and IL-12 from

mononuclear phagocytes^{11,13,14}, and IFN- γ from natural killer (NK) cells¹³ and major histocompatibility complex (MHC) class I-restricted allergen-specific CD8⁺ T cells^{6,15}, all contribute to the downregulation of IgE production *via* selection for the growth of Th1 cells. By contrast, IL-13 from CD4⁺ T cells¹⁶⁻¹⁸ acts in a similar fashion to IL-4 in promoting IgE synthesis by B cells, and this pathway is further amplified *via* the Th1-suppressive effects of CD4⁺ T-cell-derived IL-10 (Ref. 19). Transforming growth factor β (TGF- β) may also contribute indirectly to selection against Th2-cell proliferation by amplifying the production of cytokines by CD8⁺ T cells (notably IFN- γ) (Ref. 20). The circumstances that determine the relative importance of these cytokine signals in individual allergen-specific immune responses *in vivo*, particularly in humans, remain to be determined.

Animal models of 'natural' exposure to environmental allergens

It is generally accepted that immune responses (including IgE) to antigens impinging on the mucosal surfaces of the body are, to a significant extent, under local regional control. While the full range of cytokines listed above undoubtedly play significant roles in local regulation of IgE responses, an understanding of precisely how this occurs *in vivo* requires detailed analysis of the responses in the context of the tissues in which they take place.

The mucosal surfaces of principal interest in the context of atopic disease are the gastrointestinal tract (GI tract) and, in particular, the respiratory tract. The immune system at both sites is faced with comparable problems of discrimination between ubiquitous non-pathogenic antigens that should be ignored (e.g. dietary proteins, pollens), and antigens associated with microbial pathogens that merit a rapid and vigorous immune response. How this balancing act is achieved is at least partially understood. In both organ systems, the 'default' immune response to intrinsically inert soluble protein antigens that are presented to intact mucosal surfaces is the generation of a state of antigen-specific hyporesponsiveness, which was originally described as a form of immunological tolerance. This idea dates to the original observations of H.G. Wells on the induction of 'oral tolerance' by controlled feeding of egg proteins to guinea-pigs²¹, as well as to the comparatively recent description of a similar phenomenon in the respiratory tract²².

It is now clear that the essential elements of such tolerance phenomena are similar in both the GI tract and the respiratory tract. IgE and delayed-type hypersensitivity (DTH) responses are the mechanisms that are most-readily tolerized and the resulting hyporesponsiveness is the most stable. Other manifestations of host immunity (such as IgG and IgA production) are preserved to variable degrees in tolerized animals from different genetic backgrounds²³⁻²⁵. Furthermore, it is noteworthy that, during chronic exposure to inhaled antigen, the onset of tolerance in the IgE antibody isotype is preceded by a period of active IgE production^{22,26}; transient immediate hypersensitivity symptoms indicative of IgE have also been described

as a hallmark of the early phase of induction of oral tolerance²¹.

Susceptibility to tolerogenesis *via* the lung and the GI tract is genetically determined: high sensitivity being co-inherited with low-IgE-responder phenotype^{6,27,28}. However, environmental factors (particularly those associated with local mucosal inflammation) are capable of overriding this mechanism, thus converting what is normally a tolerogenic signal into a powerful immunogenic stimulus^{23-25,28}. It has been argued that this provides one avenue for the mucosal immune system to bypass local suppression and mobilize host defences against incoming microbial antigens, which are then revealed as 'pathogenic' *via* the inflammatory responses they induce^{25,29}. An illustration of this latter process was recently provided in experiments employing a strain of *E. coli* expressing transfected ovalbumin (OVA) genes: oral administration of native OVA to rats induced OVA-specific oral tolerance, whereas feeding the transfected organism induced vigorous anti-OVA immunity³⁰. While this bypass mechanism may serve an important function in host defence against pathogens, it may also provide a potential window for primary allergic sensitization to bystander environmental antigens that are encountered during inflammatory responses to microbial agents or chemical irritants^{24,25}. This conclusion is consistent with a significant body of epidemiological literature on the etiology of allergic disease in humans²⁹.

The nature of the cellular and molecular mechanisms underlying the induction of tolerance at mucosal surfaces is under intensive investigation in many laboratories. Interest has recently been fuelled by proposals that these natural processes may be amenable to exploitation in the context of therapeutic strategies, notably for suppression of ongoing pathogenic immune responses both in autoimmune and allergic diseases^{9,31-33}.

In many of the reported models, mucosally induced tolerance can be adoptively transferred with purified T cells from tolerized animals, in particular those expressing the CD8⁺ phenotype^{24,25}. Two recent studies have demonstrated inhibition of such transfer by using blocking antibodies against IFN- γ and TGF- β (Refs 34,35). Both feeding and inhalation of soluble protein antigens stimulate high levels of IFN- γ production by T cells from the regional lymph nodes that drain the respective mucosal surfaces during the phase preceding tolerance induction^{6,36}. This includes a major component of allergen-specific CD8⁺ T cells that have been suggested to 'select' for CD4⁺ Th1 cells of the same specificity with continued allergen exposure⁶. Furthermore, it can be suggested that 'tolerance' is a misnomer for this process, particularly in view of the fact that it is characterized by suppression of some manifestations of immunity (i.e. immediate hypersensitivity and DTH) with the simultaneous preservation of others (notably IgG and IFN- γ -producing T-cell reactivity). Indeed, it may be more-accurately described as 'immune deviation'⁶.

The efficiency of immune deviation is much higher in immunologically naive animals than in those with established T-cell memory. Thus, exposure to inhaled

allergen at the low levels normally encountered in the environment is not only unable to 'turn off' ongoing IgE responses in primed animals, but also actively boosts them²⁵, presumably by stimulating existing allergen-specific Th2 memory cells. Accordingly, it is reasonable to postulate that if a similar set of mechanisms contributes to downregulation of IgE responses to environmental allergens in humans, they are most likely to exert their major effects at, or around the time of, initial exposure to the allergens. In most cases, this equates to infancy or early childhood.

Natural history of IgE responses to environmental allergens in humans

Until comparatively recently, the hyperreactivity of atopic individuals to environmental allergens was ascribed principally to a generalized failure of protective 'barrier' mechanisms that normally prevent effective interaction between exogenous allergens and the immune system. This notion has been steadily eroded by recent findings (summarized below) on the widespread nature of IgG antibody responsiveness to these antigens within the non-atopic population, and was finally put to rest by the discovery that the peripheral blood of normal individuals contains CD4⁺ T cells that are specific for inhalant (aero)allergens. These cells give rise to clones that typically exhibit a Th1-like cytokine profile, in contrast to atopic individuals, whose profile resembles the murine Th2 pattern³⁷⁻³⁹. This argues strongly that active ongoing 'recognition' of environmental allergens is virtually universal, and that the feature distinguishing the allergen-responder status of atopic individuals from that of non-atopic individuals is the nature of the T-cell subsets that dominate their respective allergen-specific T-memory pools. Once a stable memory population is established for a particular allergen, it could theoretically 'lock' the host CD4⁺ T-cell response into a permanent Th1-like or Th2-like pattern by virtue of the stimulation provided *via* the continuous trickle of incoming allergen from the natural environment. Such a scenario could account both for the progressively increasing intensity of the allergic response often seen in the Th2-reactive atopic individuals, and the (usually) permanent resistance of Th1-reactive non-atopic individuals to the development of allergy.

When is T-cell memory for environmental allergens initially established in humans? The available evidence (reviewed in Ref. 29) points to early childhood. It is clear from recent studies employing sensitive antibody assays that almost all children initiate IgG responses against common aeroallergens by three months of age, and that these are almost exclusively of the IgG1 subclass⁴⁰. These responses persist throughout childhood into adulthood, by which time they are usually accompanied by a parallel IgG4 response^{41,42}. IgG1 production against common food allergens is also universal amongst infants and (unlike the response to aeroallergens) is accompanied by a specific IgG4 component from the outset⁴². It is of interest to note that these food-allergen-specific IgG1 responses peak both in affinity and titre during the neonatal period and wane thereafter with continued stimulation,

suggesting the onset of some form of antigen-specific downregulation⁴².

This biphasic pattern of responsiveness is more marked with respect to IgE antibodies to the same allergens. Long-term prospective studies employing sensitive low-level antibody assays indicate that the appearance of food-allergen-specific IgE is common both in atopic and normal infants, peaking before one year of age in the majority of cases; these responses attain higher titres in the atopic population, but decline in most children to undetectable levels by age 2-3 years^{43,44}. The levels attained in normal children are generally well below those associated with clinical symptoms, whereas a subset of the atopic children develop antibody titres that are potentially pathological⁴³.

IgE responses to aeroallergens follow a similar pattern, except that the time frame is markedly different. The production of IgE to these allergens is again common both in atopic and non-atopic children, but is usually not detected until after one year of age⁴³⁻⁴⁵. The biphasic pattern observed with respect to food-allergen-specific IgE responses is repeated, but is spread over a considerably longer period. Peak levels in atopic children are commonly not attained until four years of age, and may take an equivalent further period to decline to below the limits of detection. By contrast, IgE antibody titres in atopic children typically rise more rapidly and peak at a higher level, and approximately 30% of this group maintain high IgE titres to one or more allergens beyond the age of 12 years^{43,44}. It is noteworthy in this context that transient skin-test reactivity to food antigens occurs commonly in infants during the first year of life, whereas reactions to inhalant allergens are much less frequent, develop later in childhood, and often persist and increase with age⁴⁵.

T-cell regulation of IgE responses to environmental allergens in non-atopic humans

The epidemiological information concerning the known pattern of allergen-specific CD4⁺ T-cell reactivity in adult humans, together with the available data from the literature on exposure of experimental animals to mucosal allergens, suggest the following model for the etiology of human allergic disease. During early childhood, encounters with ubiquitous food and airborne allergens trigger CD4⁺ T-cell responses of varying intensity in all individuals. This initially involves Th0-like, Th1-like and Th2-like elements, and leads to heterogeneous allergen-specific antibody responses that includes low-level IgE production (Fig. 1). As a result of continued stimulation from these ubiquitous environmental allergens, an antigen-driven selection process ensues within each immune response, involving cross-regulation of competing Th1 and Th2 cells. In non-atopic individuals, this leads to the eventual emergence of a dominant Th1 memory population and the concomitant waning of Th2-dependent IgE production. By contrast, for reasons that are currently unclear, this natural antigen-driven selection process occasionally fails, leading instead to selection for stable Th2 reactivity, and eventually atopy. The apparent restriction of propensity for CD4⁺ Th2 'hyperreactivity' in atopic individuals to environmental (especially inhaled)

allergens, as opposed to antigens in general³⁷, suggests that their failure to downregulate IgE responses adequately stems from a malfunction in local mucosal immune regulation, as opposed to a primary defect in T-cell function.

It is particularly interesting that, even amongst atopic individuals, IgE-mediated food allergy is extremely rare, and this is reflected in the high efficiency with which IgE responses to food antigens are 'switched off' during early childhood. Indeed, close to 100% of atopic children lose their IgE reactivity to foods by the age of four years, whereas at least 30% maintain significant titres to one or more aeroallergens^{43,44}.

A summary of the salient features of the 'normal' IgE response to ubiquitous environmental allergens during childhood is shown in Table 1. This raises the question as to why the downregulation of IgE is more rapid and effective for responses to ingested allergens than to inhaled allergens. While intrinsic differences in local antigen processing/presentation may play a role, the emerging picture of local antigen-presenting cell (APC) populations in the two organ systems show striking similarities. This is particularly true with respect to the dynamics and functional phenotype of the resident dendritic cell populations that appear to regulate primary immunity to soluble protein antigens in the steady state⁴⁶⁻⁴⁸. A more-feasible explanation would appear to be the relative intensity of exposure to local allergens at the two sites. Thus, the higher concentrations of dietary antigen may drive this antigen-specific T-cell-selection process much faster and more efficiently in the gut-associated lymphoid tissues than the parallel process in the lung, where the stimulus is represented by intermittent exposure to nanogram levels of airborne antigen. In this context, it has recently been demonstrated that, whereas feeding low levels of exogenous antigen (in the microgram range) induces T-cell-mediated suppression or immune deviation, feeding higher levels of the same antigens (in the milligram range) produces T-cell 'anergy', which cannot be adoptively transferred⁴⁹. It is also of interest to note that high levels of IFN- γ secretion are induced by high-dose feeding³⁶, suggesting that allergen-specific cells that evade the anergizing process would most likely be of the Th1 phenotype. As suggested in Table 1, the operation of these complementary mechanisms in concert in the GI tract may account for the high efficiency of selection against food-allergen-specific Th2 immunity relative to the parallel process in the respiratory tract, where low-level exposure to inhalant allergen may normally be sufficient to stimulate only the mechanisms of immune deviation.

Quantitative differences in the levels of allergen stimulation in the two organ systems may also underlie the variations in the kinetics of IgG4 responses noted above. Thus, the early postnatal IgG response to food, but not inhalant allergens, typically contains an IgG4 component that, in other systems, is a direct reflection of repeated intense antigenic stimulation^{50,51}.

Is intervention feasible?

It is important to reiterate two salient facts before considering the question of therapeutic intervention.

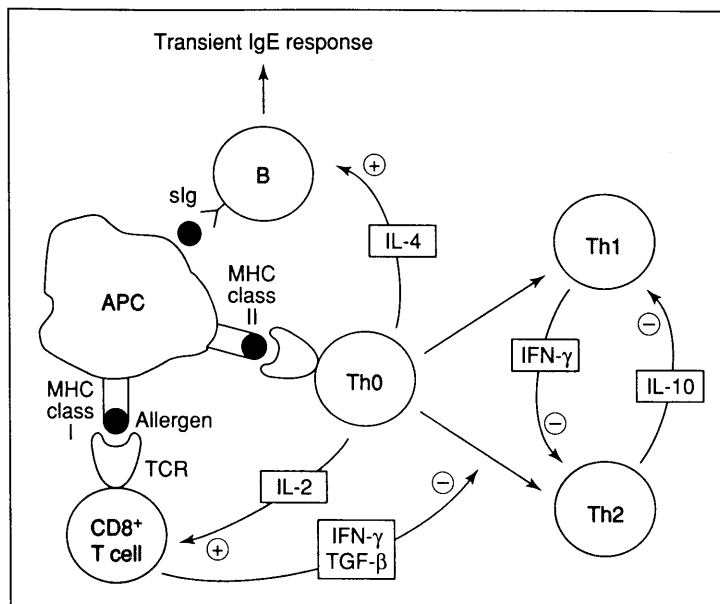


Fig. 1. Selection for protective T helper 1 (Th1)-mediated cell immunity to allergens during the primary immune response via major histocompatibility complex (MHC) class I-restricted CD8⁺ T-cell-mediated immune deviation. Differences in the shading of allergenic peptides reflect the presence of different peptides bound to MHC class I or class II, which are presented to CD8⁺ T cells or Th0 cells, respectively. Abbreviations: APC, antigen-presenting cell; IL-4, interleukin 4; IFN- γ , interferon γ ; TGF- β , transforming growth factor β ; slg, surface immunoglobulin; TCR, T-cell receptor.

First, despite the high frequency and wide distribution of potential aeroallergens in the natural environment, atopic individuals usually display allergic reactivity to a limited spectrum of allergens, to the extent that single specificities have been shown to account for greater than 50% of total serum IgE antibody in some patients⁵². This suggests that, even in these susceptible individuals, the natural mechanism(s) that selects against Th2 reactivity to inhaled allergens functions effectively for most allergens. Second, the occurrence of high-titre food-antigen-specific IgE responses in adults is extremely rare, reflecting the high success rate (almost 100%) achieved for termination of food-antigen-specific IgE responses during infancy⁴⁴.

Table 1. Features of 'normal' IgE responses to environmental allergens during early childhood

	Inhalant allergens	Food allergens
Exposure levels	ng	μ g-g
Initial appearance of serum IgE	1-2 years	3-6 months
Duration of 'transient' IgE responses	3-5 years	1-2 years
Skin-test reactivity	- ^a	+ ^b
Clinical sequelae	?	+
Likely mechanism(s) for Th1 'selection'	Immune deviation	Immune deviation, T-cell anergy

Abbreviations: Th1, T helper 1.

^aPositive responses essentially restricted to atopic children.

^bTransient response.

Accordingly, this protective mechanism should be viewed overall as highly efficient in the respiratory tract, and approaching perfection in the GI tract, at least with respect to IgE regulation. However, the natural 'oral tolerance' process may have a downside, particularly during the heterogeneous phase of the host response to food antigens in early childhood: pediatricians and parents alike recognize this as a transient period of often frequent and unexplained gastroenteric symptoms, and it is likely that the immuno-inflammatory phenomena described above contribute to this situation. Nonspecific respiratory symptoms are also a feature of early childhood, but are typically less frequent and less severe, and this may be a reflection of the lower intensity of exposure to inhalant allergen.

In relation to the etiology of atopic disease, it can be argued logically that if the efficiency of the natural process that terminates IgE responses to inhaled allergens during childhood could be improved to approach that of the parallel process that successfully regulates responses to food allergens, a substantial proportion of the burden of allergic respiratory disease in the community would be removed. Is this a realistic possibility? First principles suggest that it may be. The general approach to be followed appears (on paper) relatively straightforward: interpose appropriate allergen-specific signals into the natural T-cell selection process during the early stages of infancy and, thereby, tip the equilibrium towards selection for Th1-like immunity. This may be achievable by the simple device of repeated feeding with aeroallergens in order to stimulate the tempo of this normally sluggish selection process (relative to the process in the GI tract). In reality, this would involve administration of an oral 'vaccine' comprising a cocktail of natural or recombinant antigens. Theoretically, such mixtures need only contain a small number of major aeroallergens to be effective, owing to two principal reasons: (1) a limited spectrum of allergens appears to be responsible for the bulk of allergic respiratory disease in most geographic regions; and (2) cytokine-mediated bystander effects are likely to contribute to the process of oral tolerance^{53,54}. The latter may also render feasible the use of allergen-derived synthetic peptides covering only a few immunodominant epitopes, given that the multiple specificities characteristic of major aeroallergens such as house dust mite (HDM) are invariably presented together at mucosal surfaces in their native form as a mixture. The potential efficacy of this approach has recently been demonstrated *via* the successful induction of oral tolerance to intact Der p1 allergen from HDM by feeding with a single T-cell epitope⁵⁵.

It may be possible to refine this general approach by using other mucosal routes for controlled allergen administration, notably intranasal^{22,55} or sublingual⁵⁶, which require log-scale lower amounts of allergen to achieve the same results as allergen feeding. A further refinement for consideration is the use of parenteral immunization regimes, exploiting some of the emerging vaccine strategies for selective stimulation of MHC class I-restricted pathways^{57,58}, and hence biasing ensuing T-cell responses towards IFN- γ .

Limitations of the approach

Two critical ethical issues must be addressed before embarking on the intervention approach described above, notably: (1) what are the possible side-effects of interfering with the natural process of 'selection' of CD4⁺ T-cell memory for inhalant allergens in children who (at least at the time of 'vaccination') are free of respiratory allergy; and (2) precisely how is the potential target population to be selected?

With respect to the former, the overriding concern must be the potential for induction of classical DTH. Currently, there is no convincing evidence for a significant DTH component in asthma or rhinitis, but inadvertent 'priming' with inhalant allergens under the artificial circumstances referred to above could theoretically alter this situation. However, DTH correlates with IgE at the high end of the spectrum of *in vivo* sensitivity to induction of mucosal tolerance. In addition, attempts in many laboratories to develop models of chronic tissue damage in the respiratory tract mediated by DTH in response to inhalation of soluble protein antigens have failed. Although more-artificial systems, employing intratracheal administration of a liquid bolus containing contact-sensitizing agents⁵⁹ or pro-inflammatory microbial extracts of cell-wall products from organisms such as *Micropolyspora faeni*⁶⁰ and *Thermoactinomyces*⁶¹, have proved more successful, these agents share little in common with natural exposure to intrinsically inert aeroallergens. Thus, it is clear that more research is required in this area.

The issue of defining the potential target population for vaccination against aeroallergens balances on the question of genetic risk. Although current opinion is divided on the precise nature of inheritance patterns in atopy, there is general agreement that high-risk individuals can be readily identified on the basis of family history: the degree of risk rising with the number of first-degree relatives manifesting the disease. More-objective methods for identification of predisposed individuals may also soon be available through ongoing developments in molecular genetics. This is exemplified by recent studies that have identified a putative 'atopy gene' on chromosome 11q, which appears to be a marker for high IgE production in a subset of allergy-prone families⁶².

In summary, this article has argued that the key to the control of allergic disease may lie in exploiting endogenous immunological mechanisms that normally protect mucosal surfaces against the consequences of inappropriate responses to non-pathogenic soluble protein antigens. While acknowledging the imperfect nature of our understanding of precisely how these mechanisms operate *in vivo*, the basic principles underlying the model proposed above appear entirely consistent with current views of the role of crossregulation between T-cell subsets in the determination of qualitative and quantitative aspects of the immune response. Indeed, the unresolved issues that currently stand in the way of testing the general immunoprophylactic strategy suggested here appear minor in comparison with those currently being addressed in research programs that focus on regulation of individual steps in the complex molecular cascade underlying the expression

of atopic disease. Diversion of part of this latter research effort towards development of a Th1-selective 'vaccine' for use initially with the major aeroallergens would appear amply justified on the basis of current knowledge.

P.G.H. is supported by the National Health and Medical Research Council of Australia.

Patrick G. Holt is at the Institute for Child Health Research, PO Box 855, West Perth, WA 6872, Australia.

References

- 1 Sears, M., Burrows, B., Flannery, E., Herbison, G., Hewitt, C. and Holdaway, M. (1991) *New Engl. J. Med.* 325, 1067-1071
- 2 Bauman, A., Lyle, D., Taylor, L. and Thomson, P. (1990) *Aust. Med. Rec. J.* Sept., 101-105
- 3 Weiss, K.B., Gergen, P.J. and Hodgson, T.A. (1992) *New Engl. J. Med.* 326, 862-866
- 4 Sullivan, S.D. and Weiss, K.B. (1993) *Allergy* 48, 146-152
- 5 Finkelman, F.D., Holmes, J., Katona, I.M. *et al.* (1990) *Annu. Rev. Immunol.* 8, 303-333
- 6 McMenamin, C. and Holt, P.G. (1993) *J. Exp. Med.* 178, 889-899
- 7 Mosmann, T.R. and Coffman, R.L. (1989) *Annu. Rev. Immunol.* 7, 145-173
- 8 Romagnani, S. (1991) *Immunol. Today* 12, 256-257
- 9 O'Hehir, R.E., Hoyne, G.F., Thomas, W.R. and Lamb, J.R. (1993) *Eur. J. Clin. Invest.* 23, 763-772
- 10 Robinson, D.S., Hamid, Q., Ying, S. *et al.* (1992) *New Engl. J. Med.* 326, 298-304
- 11 Pène, J., Rousset, F., Brière, F. *et al.* (1988) *Proc. Natl Acad. Sci. USA* 85, 6880-6884
- 12 Lundgren, M., Persson, U., Larsson, P. *et al.* (1989) *Eur. J. Immunol.* 16, 1311-1315
- 13 Romagnani, S. (1992) *Immunol. Today* 13, 379-381
- 14 Manetti, R., Parronchi, P., Giudizi, M.G. *et al.* (1993) *J. Exp. Med.* 177, 1199-1204
- 15 Kemeny, D.M. and Diaz-Sanchez, D. (1993) *Clin. Exp. Allergy* 23, 466-470
- 16 McKenzie, A.N., Culpepper, J.A., de Waal Malefyt, R. *et al.* (1993) *Proc. Natl Acad. Sci. USA* 90, 3735-3739
- 17 Minty, A., Chalon, P., Derocq, J.-M. *et al.* (1993) *Nature* 362, 248-250
- 18 Punnonen, J., Aversa, G., Cocks, B.G. *et al.* (1993) *Proc. Natl Acad. Sci. USA* 90, 3730-3734
- 19 Mosmann, T.R. and Moore, K.W. (1991) *Immunol. Today* 12, A49-A53
- 20 Lee, H.M. and Rich, S. (1993) *J. Immunol.* 151, 668-677
- 21 Wells, H.G. and Osborne, T.B. (1911) *J. Infect. Dis.* 8, 66-124
- 22 Holt, P.G., Batty, J.E. and Turner, K.J. (1981) *Immunology* 42, 409-417
- 23 Holt, P.G. and Sedgwick, J.D. (1987) *Immunol. Today* 8, 14-15
- 24 Mowat, A.M. (1987) *Immunol. Today* 8, 93-98
- 25 Holt, P.G. and McMenamin, C. (1989) *Clin. Exp. Allergy* 19, 255-262
- 26 Sedgwick, J.D. and Holt, P.G. (1985) *Cell. Immunol.* 94, 182-194
- 27 Sedgwick, J.D. and Holt, P.G. (1984) *Eur. J. Immunol.* 14, 893-897
- 28 Holt, P.G., Britten, D. and Sedgwick, J.D. (1987) *Immunology* 60, 97-102
- 29 Holt, P.G., McMenamin, C. and Nelson, D. (1990) *Pediatr. Allergy Immunol.* 1, 3-13
- 30 Dahlgren, U.I., Wold, A.E., Hanson, L.A. and Midtvedt, T. (1991) *Immunology* 73, 394-397
- 31 Metzler, B. and Wraith, D.C. (1993) *Int. Immunol.* 5, 1159-1165
- 32 Nussenblatt, R.B., Caspi, R.R., Mahdi, R. *et al.* (1990) *J. Immunol.* 144, 1689-1695
- 33 Weiner, H.L., Mackin, G.A., Matsui, M. *et al.* (1993) *Science* 259, 1321-1324
- 34 Zhang, Z. and Michael, J.G. (1990) *J. Immunol.* 144, 4163-4165
- 35 Miller, A., Lider, O., Roberts, A.B. and Sporn, M.B. (1992) *Proc. Natl Acad. Sci. USA* 89, 421-425
- 36 Hoyne, G.F., Callow, M.G., Kuhlman, J. and Thomas, W.R. (1993) *Immunology* 78, 534-540
- 37 O'Hehir, R.E., Bal, V., Quint, D. *et al.* (1989) *Immunology* 66, 499-504
- 38 Wierenga, E.A., Snoek, M., de Groot, C. *et al.* (1990) *J. Immunol.* 144, 4651-4656
- 39 Field, E.H., Noelle, R.J., Rouse, T., Goeken, J. and Waldschmidt, T. (1993) *J. Immunol.* 151, 48-59
- 40 Mariani, F., Price, J.F. and Kemeny, D.M. (1992) *Clin. Exp. Allergy* 22, 29-33
- 41 Kemeny, D., Urbanek, R. and Ewan, P. (1989) *Clin. Exp. Allergy* 19, 545-549
- 42 Devey, M.E., Beckman, S. and Kemeny, D.M. (1993) *Clin. Exp. Immunol.* 94, 117-121
- 43 Hattevig, G., Kjellman, B. and Björkstén, B. (1987) *Clin. Allergy* 17, 571-578
- 44 Hattevig, G., Kjellman, B. and Björkstén, B. (1993) *Pediatr. Allergy Immunol.* 4, 182-186
- 45 Van Asperen, P.P. and Kemp, A.S. (1989) *Acta Paediatr. Scand.* 78, 239-245
- 46 Holt, P.G., Schon-Hegrad, M.A. and Oliver, J. (1988) *J. Exp. Med.* 167, 262-274
- 47 Schon-Hegrad, M.A., Oliver, J., McMenamin, P.G. and Holt, P.G. (1991) *J. Exp. Med.* 173, 1345-1356
- 48 Liu, L.M. and MacPherson, G.G. (1993) *J. Exp. Med.* 177, 1299-1307
- 49 Gregerson, D.S., Obritsch, W.F. and Donoso, L.A. (1993) *J. Immunol.* 151, 5751-5761
- 50 Aalberse, R.C., van der Gaag, R. and van Leeuwen, J. (1983) *J. Immunol.* 130, 722-726
- 51 Urbanek, R., Kemeny, D.M. and Richards, D. (1986) *Clin. Allergy* 16, 317-322
- 52 Chapman, M.D. and Platts-Mills, T.A.E. (1980) *J. Immunol.* 125, 587-592
- 53 Vaz, N.M., Maia, L.C., Hanson, D.G. and Lynch, J.M. (1981) *Mem. Ist. Oswaldo Cruz* 76, 83-91
- 54 Miller, A., Lider, O. and Weiner, H.L. (1991) *J. Exp. Med.* 174, 791-798
- 55 Hoyne, G.F., O'Hehir, R.E., Wraith, D.C., Thomas, W.R. and Lamb, J.R. (1993) *J. Exp. Med.* 178, 1783-1788
- 56 Holt, P.G., Vines, J. and Britten, D. (1988) *Clin. Allergy* 18, 229-234
- 57 Collins, D.S., Findlay, K. and Harding, C.V. (1992) *J. Immunol.* 148, 3336-3341
- 58 Martinon, F., Gras Masse, H., Boutillon, C. *et al.* (1992) *J. Immunol.* 149, 3416-3422
- 59 Garssen, J., Nijkamp, F.P., Wagenaar, S.S., Zwart, A., Askenase, P.W. and Van Loveren, H. (1989) *Immunology* 68, 51-58
- 60 Schuyler, M., Cook, C., Listrom, M. and Fengolio, P.C. (1988) *Am. Rev. Respir. Dis.* 137, 1449-1455
- 61 Takizawa, H., Suko, M., Kobayashi, N. *et al.* (1988) *J. Allergy Clin. Immunol.* 81, 391-400
- 62 Sandford, A.J., Shirakawa, T., Moffatt, M.F. *et al.* (1993) *Lancet* 341, 332-334