

Review

Environmental factors and primary T-cell sensitisation to inhalant allergens in infancy: reappraisal of the role of infections and air pollution

Holt PG. Environmental factors and primary T-cell sensitisation to inhalant allergens in infancy: reappraisal of the role of infections and air pollution.

Pediatr Allergy Immunol 1995; 6: 1–10. © Munksgaard 1995.

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Received September 1, 1994

Accepted September 23, 1994

There is increasing evidence that both the prevalence and severity of atopic diseases, in particular those affecting the respiratory system, are progressively increasing in the developed countries (1), and similar trends are evident in traditionally rural societies which are in the process of urbanisation (2). If allergy is really a malaise of "modern society", what are the underlying societal/environmental factors responsible for its emergence?

The thesis advanced in the review below argues that:

a) primary sensitisation to inhalant allergens results from failures in cognate allergen-specific "selection" processes, that normally set protective patterns of the TH-1-like immunological memory to environmental antigens during infancy;

b) infancy is a "high risk" period for such failures, due to intrinsic maturational deficiencies in T-cell-dependent and -independent immunoinflammatory function(s);

c) the principal impetus for normal postnatal maturation of immunoinflammatory function(s) is provided by microbial stimulation, from both normal commensals and pathogens in the gastrointestinal and respiratory tracts;

d) major improvements in public health and personal hygiene practices, changes in infectious disease management (especially the widespread use of antibiotics in pediatrics), reductions in average family sized, and improvements in general living standards in the developed countries over the last 2–3 generations, have collectively lowered the overall level of exposure to microbial stimulation during infancy, resulting (at a population level) in slower postnatal maturation of the immune system and thus prolongation of the "high risk" period for primary allergic sensitisation.

Recent epidemiological findings, linking high frequency of childhood respiratory infections in populations with low socio-economic status with reduced prevalence of allergic sensitisation, appear to support this view. It is further argued that the cellular regulatory mechanism(s) set in motion by high levels of microbial stimulation, which normally skew T-cell responses to environmental allergens towards TH-1, are sufficiently powerful to override the potentially "pro-sensitisation" effects of domestic and industrial air pollution.

T-cell regulation of IgE production: cellular and molecular mechanism(s)

Experimental animal models

In vitro – A single development in basic immunology has shaped the last decade of research in the allergy field, notably the concept of partitioning of cytokine responses within discrete subpopulations of CD4⁺ T-helper cells (review in 3, 4). This concept evolved initially from the study of T-cell clones derived from animals mounting immune responses against microbial infections and parasites, but was subsequently found to be equally applicable to immune responses against non-pathogenic soluble protein antigens including potential “allergens”.

The key cytokine in relation to allergic disease is IL-4, which instructs uncommitted B-lymphocytes to “switch” to IgG₁ and IgE production (5, 6). IL-4 is produced by CD4⁺ T-cells of the T-helper-2 (TH-2) subset, which compete with antagonistic T-helper-1 (TH-1) cells for “dominance” of primary immune responses; at the clonal level, this competition involves secretion of the TH-1-inhibitory cytokine IL-10 by TH-2 cells, versus secretion of TH-2-inhibitory IFN γ by TH-1 cells (3, 4).

These findings have given rise to a general model for IgE immunoregulation which now enjoys wide acceptance, viz., that the magnitude of the IgE component of antigen specific immune responses to persistent antigens is ultimately determined by the relative balance between antigen-specific TH-1 and TH-2 cells that becomes established in relevant T-memory populations. It is further generally accepted that at a population level, the principal selection pressures that determine this balance are cytokines produced by a range of bone marrow-derived cells (5, 6).

In vivo experimental models – The experimental models of principal relevance to our understanding of the aetiology of allergic diseases, are those which have focussed upon the nature of immune responses to soluble protein antigens presented under “natural” conditions of passive exposure, at mucosal surfaces. In particular, these involve exposure to antigens via feeding and aerosol inhalation.

The history of experiments on the induction of allergy to dietary antigens dates back to the pioneering studies of Wells in 1911 (7) involving the feeding of egg proteins to guinea pigs: after an initial period of clinical hypersensitivity (which included some deaths by anaphylaxis!), the surviving animals in which exposure was continued developed resistance to further challenge. Subsequent studies confirmed the existence of this “oral tolerance” phenomenon and demonstrated its selectivity for IgE, and moreover showed that transient IgE production commonly preceded the onset of tolerance (8).

The cellular mechanism(s) mediating oral tolerance have been controversial, due to the variable success reported in the ability to “transfer” tolerance to naive animals employing purified T-cells from tolerant donors (8). However, recent studies have served to reconcile these differences, and have demonstrated that two regulatory mechanisms for oral tolerance co-exist, and their relative importance varies with antigen dose (9).

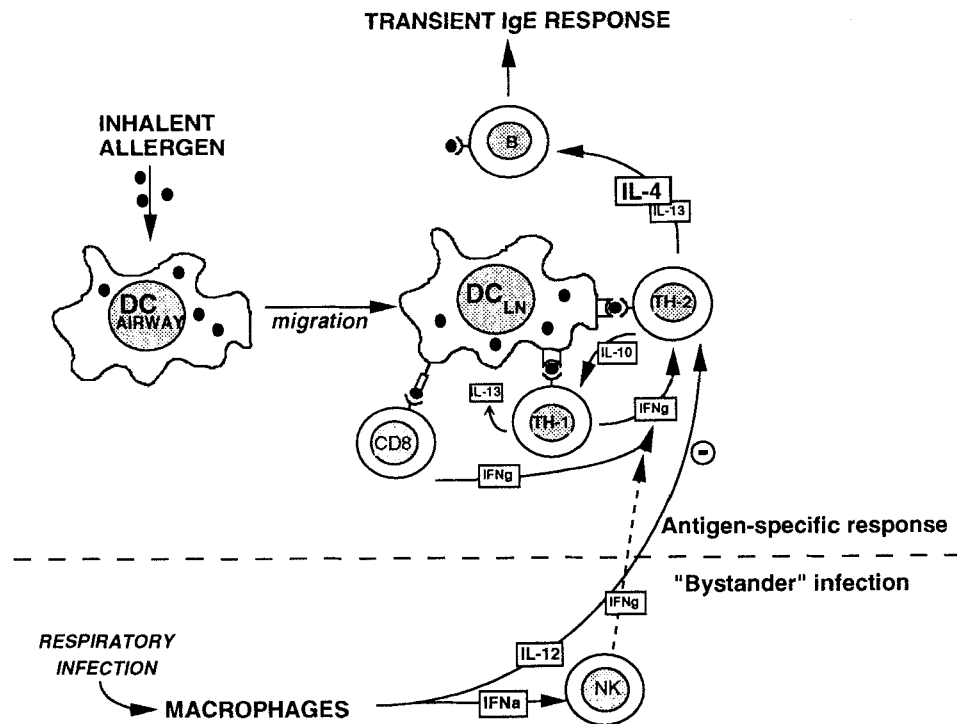
Thus, when feeding high levels of antigen, the dominant mechanism involves functional deletion of food antigen-specific T-cells via direct T-cell anergy, whereas when tolerance is induced by longer-term exposure to low levels of food antigen, the dominant mechanism involves “suppression” by antigen-specific T-cells (9) of the classical “suppressor” CD8⁺ phenotype (10–12). At mid-points in the dose curve, it is likely that both mechanisms can operate side-by-side; it is also of interest to note that during feeding of high doses of antigen, high levels of IFN γ are produced by T-cells from lymph nodes draining the GIT (13), which suggests that any food-antigen-specific CD4⁺ T-cells which “escaped” the anergy process would tend to be of the TH-1 (i.e. IFN γ -resistant) phenotype.

Detailed studies from our lab have provided new insight into the parallel process in the respiratory tract, in response to exposure to “physiological” (viz. very low) levels of inhaled allergen. Our earlier work (review 14) indicated that a similar form of antigen-specific tolerance normally developed in the IgE antibody class after repeated inhalation of antigens, which could be transferred to naive animals via CD8 T-cells.

Susceptibility to this form of tolerance induction is genetically determined (15), but can be markedly influenced by a range of environmental factors. In particular, inflammatory stimuli targetted at the airway mucosa can function as “adjuvants” to promote IgE responses to inhaled antigen (14, 16), and similar findings have been reported in relation to GIT inflammation and failure of oral tolerance mechanisms (8). In the respiratory tract, these stimuli include chemical air pollutants, and also infection (14, 16); paradoxically, however, raising experimental animals in an artificial “specified pathogen free” environment isolated from common environmental commensals/pathogens, appears to reduce the efficiency of their tolerance induction mechanism(s) (16), and this issue is addressed further below. It is also clear that critical elements of the tolerance-induction process are not fully functional at birth, as both oral tolerance and its respiratory tract equivalent fail to operate effectively during early post-natal life (review 14).

It is also important to note that it is only possible to induce tolerance in animals via inhalation or in-

Fig. 1. Primary immune response to inhaled allergens: T-helper cell selection by cytokine-mediated "Immune Deviation".



gestion, against antigens to which they are immunologically "naive" – once stable immunological memory is established in the $CD4^+$ TH-2 population and persistent IgE responses develop, further exposure to the relevant antigens only further boosts the ongoing responses (12). Additionally, this form of tolerance is preferentially directed against IgE, as "tolerised" animals typically manifest mid-range IgG responses against the same allergens i.e. the immune response is preferentially directed away from TH-2-dependent IgE production, while preserving an ongoing TH-1-dependent IgG component, a process which is known as "immune deviation" (12).

Recent experiments in our lab have focussed upon the initial cellular events occurring in the regional lymph nodes (RLN) draining the respiratory tract in response to allergen inhalation, in particular those which lead to the generation of allergen-specific $CD8^+$ T-cells which cross-regulate parallel $CD4^+$ T-cell responses. The first allergen-specific T-cells which appear in the RLN following allergen inhalation are those that secrete predominantly IL-4 and IL-2, and their appearance coincides with an initial burst of allergen-specific IgE antibody synthesis, which is detectable in serum (12). This latter may be TH-2 cells, or alternatively their TH-0 precursors (5). Over the next few days, IgE synthesis is shut down (and eventually replaced by an IgG response), the decline occurring concomitant with the appearance of allergen-specific $CD8^+$ T-cells secreting high levels of IFN γ (12). These $CD8^+$ T-cells

require an external source of IL-2 for initial activation, which is provided by the TH-2 (or possibly TH-0) cells (12). This $CD8^+$ -derived IFN γ provides a strong "immune deviation" signal, which appears to play a major role in determining the outcome of the emerging $CD4^+$ TH-cell response, selecting against TH-2 cells (12) via the scheme shown in Fig. 1. The overall efficiency of this "protective" $CD8^+$ T-cell response is genetically determined, as it is clearly switched on following allergen challenge much more rapidly (12) and at lower allergen-exposure levels (15) in immunologically normal low-IgE-responder animals, than in those expressing the high-IgE-responders phenotype.

We have also shown that unlike $CD4^+$ T-cells which recognize processed allergen presented via MHC class II molecules, these $CD8^+$ T-cells are MHC class I restricted (12). Until recently, it was believed that the only foreign antigens that could gain access to class I antigen processing pathways were those derived from infectious viruses, and that all the resulting class I MHC-restricted antigen-specific $CD8^+$ T-cells were classical cytotoxic T-lymphocytes (CTLs); however, this paradigm is no longer universally applicable, as "leakage" of endocytosed soluble protein allergens into the cytosolic compartment of antigen presenting cells has been demonstrated *in vivo*, and can result in primed $CD8^+$ T-cells that respond to their antigen(s) via cytokine secretion, as opposed to cytotoxicity (see discussion in 12).

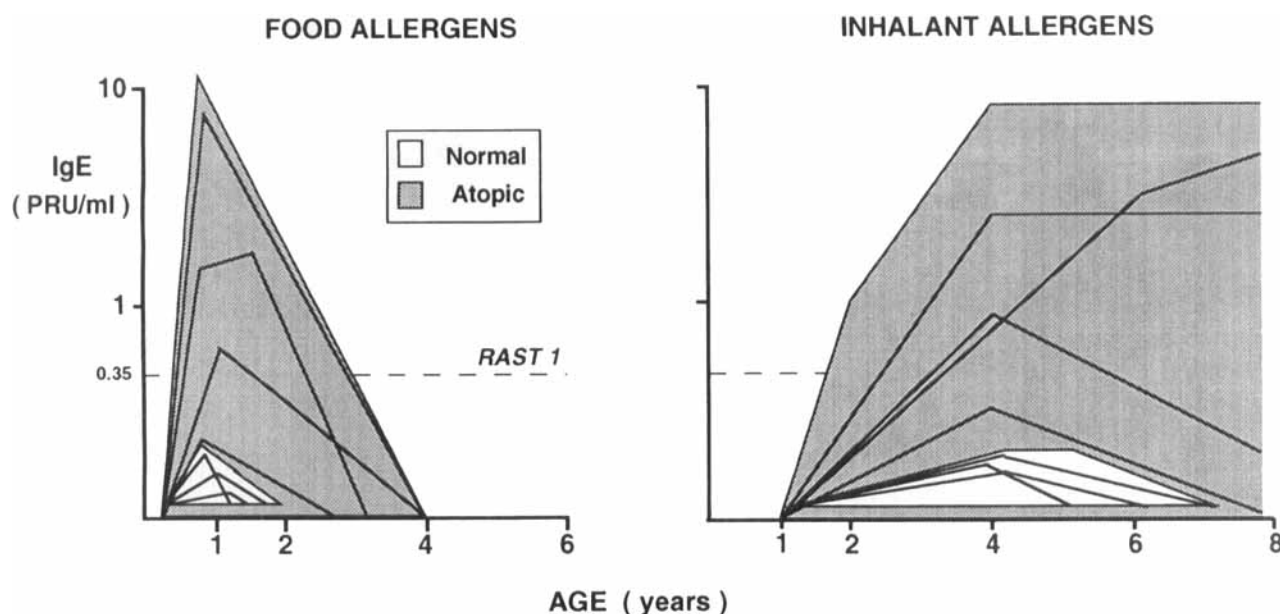


Fig. 2. Postnatal serum IgE responses to environmental allergens in normal and atopic children, derived from data in References 34 and 35. Children were bled at regular intervals over the period shown. The curves are exemplary of the types of time-dependent changes seen in antibody levels of individual children; curves for atopic children all fell within the shaded area and those for normals were restricted to the clear areas.

In addition to the contribution of IFN γ signals from CD8 $^{+}$ T-cells, the TH-1/TH-2 selection process can be influenced by cytokine signals derived from other sources external to the CD4 $^{+}$ TH-cell population. The most potent of these appear to be IL-12 and IFN α produced by mononuclear phagocytic cells in response to microbial challenge (6). These cytokines inhibit the growth of TH-2 cells directly, and/or via stimulation of IFN γ production by adjacent natural killer cells (6). Thus, as illustrated in Fig. 1, "bystander" infections in the respiratory tract which occur concomitantly with exposure to inhalant allergen could, by stimulating the migration of activated IFN α /IL-12-secreting phagocytes from infected airway tissue into the RLN, directly influence the outcome of the TH-1/TH-2 selection process occurring within the evolving allergen-specific primary immune response, favouring the outgrowth of TH-1 cells.

IgE regulation in man

In vitro – The data accumulating on cytokine regulation of IgE synthesis by human B-cells are broadly consistent with the major predictions from the animal models. Notably, *in vitro* IgE synthesis is stimulated by IL-4 (17, 18) and inhibited by IFN γ (17, 19), IFN α (19) and IL-12 (20). Certain differences have emerged, in particular concerning IL-10 which is produced by both TH-1 and TH-2 cells in man (21); this cytokine can on the one hand prevent IL-4-induced IgE synthesis by inhibiting accessory cell functions (22), but can also indirectly stimulate

IgE production via suppressing IL-12-induced IFN γ in NK cells (23). Additionally, IL-13 appears to play an important role in humans in mimicking the IgE-stimulatory effects of IL-4 (24), whereas this pathway appears relatively minor in rodents. However, the compartmentalisation and polarisation of allergen-specific CD4 $^{+}$ T-cell responses in human atopics towards a TH-2-like cytokine profile, compared to a TH-1-like profile in normals (6, 25), strongly suggests that the essential elements of this cytokine-mediated regulatory process have been conserved by evolution throughout the species.

IgE responses to inhalant allergens in humans: reactivity in adulthood is commonly "set" during early childhood – While it is clear that the *de novo* onset of "priming" to new inhalant allergen specificities can occur during adult life (26), the most common pattern observed, particularly in severe atopics, is that of a continuation of childhood patterns into adulthood. Moreover, there is increasing evidence that early postnatal exposure to "high" levels of inhalant allergens, particularly during the first three months of life, maximises the risk in genetically susceptible individuals for development of T-cell sensitisation of the type which eventually blossoms into atopic disease (27, 28).

This infers that the human immune system begins responding to ubiquitous inhalant allergens in early infancy. Two lines of evidence argue that this is the case. Firstly, *in vitro* T-cell proliferation tests have detected putative aeroallergen-specific reactivity in a subset of cord blood samples (29), and this finding has recently been independently confirmed (30),

suggesting that initial T-cell priming may in some cases occur *in utero*, although final acceptance of this possibility must await both cytokine and karyotype analyses of the responding cells. More direct evidence is provided via a number of studies (particularly 31, 32) which have clearly demonstrated inhalant (and food) allergen-specific IgG synthesis in very young infants. There are subtle differences in the kinetics of the responses to these two different types of allergens, which appear attributable (33) to differences in the absolute levels of allergenic stimulation in the respiratory tract (i.e. nanogram amount of inhalant allergens) versus GIT (milligram-gram levels of dietary proteins), and this issue is discussed in more detail below.

IgG antibodies to ubiquitous environmental antigens remain detectable in serum from the majority of both atopic and normals throughout life (31, 32). However, the picture for IgE antibodies is markedly different, as demonstrated by a series of prospective studies from Hattevig et al. (34, 35) which are summarised in Fig. 2. Prospective analysis of serum IgE titres to environmental allergens in individual children indicates that antibodies to foods are commonly detected in both groups during the first year of life, the magnitude and duration of these responses varying with IgE-responder phenotype. However, in virtually all cases, these initial food-specific IgE responses are terminated spontaneously by age 2–3 years (34, 35), leaving intact IgG₁ and IgG₄ responses to the same antigens, which persist into adulthood (31, 32). It is of interest to note that transient skin test reactivity to food allergens is also common during this period (36).

The pattern for IgE responses to inhalant allergens differs in a number of significant respects. Firstly, these responses are rarely initiated until after 1 (or more commonly 1.5–2) years of age; secondly, although the most common “curve shape” describing the responses of individual atopic and normal children to individual allergens is biphasic, it is not uncommon for the response to “see-saw” at low levels for >5–6 years (particularly in normal children) before finally shutting off permanently; and thirdly, whereas IgE responses against foods persist only rarely beyond infancy, a much larger proportion of children continue to produce IgE antibodies against one or more inhalant allergens into adulthood.

T-cell “sensitisation” to environmental allergens in infancy: a working model

Taking the alternative adult patterns of allergen-specific TH-1 versus TH-2 reactivity as the optional “end points” of this process, the following model is

proposed to describe the interactions between environmental allergens and the infant immune system:

- initial “priming” of the T-cell system occurs very early in postnatal life (in some cases, possibly *in utero*);
- during infancy, repeated environmental exposure “drives” allergen-specific T-cell responses; these are initially heterogeneous, comprising mutually antagonistic cross-regulating T-cell subsets, and (consistent with this heterogeneity) low levels of specific antibodies of several isotypes can be detected in serum during this period;
- repeated stimulation eventually “selects” for one or another of the cross-regulating T-cell subsets within each response, which becomes dominant thereafter;
- the emergence of a dominant TH-1 component in an individual response results in termination of IgE antibody synthesis, but permits on-going (low level) IgG production; theoretically, TH-1 domination of immunological “memory” for a particular allergen can effectively “lock-out” subsequent TH-2 reactivity (and thus protect against allergic sensitisation);
- conversely, selection for dominant TH-2 reactivity can lead to a progressively increasing IL-4/IL-13-driven IgE antibody response, and the eventual expression of atopic disease;
- the overall efficiency of selection for “protective” TH-1 immunity is intrinsically higher in responses to food allergens, compared to responses to inhalant allergens in the respiratory tract; this may reflect the operation of two complementary “suppressive” suppression mechanisms (T-cell anergy plus CD8-mediated immune deviation) in the GIT during oral tolerance development, whereas the lower levels of allergen stimulation in the respiratory tract restricts the selection process to immune deviation (33, 37);
- the risk of developing potentially pathogenic TH-2 immunity to inhalant allergens is exacerbated by “high level” exposure during early infancy.

TH-1/TH-2 “Selection” during infancy and the normal postnatal development of immunocompetence

The model above proposes that the basis for normal “protection” against allergy to ubiquitous antigens is an antigen-driven T-cell selection process during childhood which if successful, results in the development of dominant TH-1 “memory” for individual allergens.

The scheme in Fig. 1 envisages four cell populations as playing the principal rate-limiting roles in this process, namely class II MHC-restricted

CD4⁺ T-cells, class I MHC-restricted CD8⁺ T-cells, airway derived dendritic cells (the archetypal "antigen-presenting cell" for naive T-cells), and macrophages. The functional status of these cell populations in neonates are briefly as follows:

Macrophage and monocyte migration into sites of inflammation is delayed and attenuated in neonates, which may underlie their low skin test reactivity (38, 39); this may be partly due to diminished capacity to express surface integrins such as Mac-1 (40). Their capacity to both produce (41–43) and respond to (42) a number of cytokines is reduced relative to adults. No direct information is available on tissue macrophages from the airway wall in neonates, but those accessible by bronchoalveolar lavage are clearly functionally immature (44, 45; see also below).

CD8⁺ T-cells from human infants perform poorly in classical assay systems which measure cytotoxic effector functions (46, 47), but their cytokine responses *per se* have not been investigated.

CD4⁺ T-cell function is variably but significantly reduced in the early postnatal period, and certain key aspects do not completely mature for several years. This may be in part attributable to the variable contribution of phenotypically "foetal-type" thymocytes to the peripheral T-cell compartment in infants (48–51). Of particular note is the lowered capacity of neonatal T-cells to produce IFN γ following stimulation (45–47), and moreover the postnatal maturation of this important function appears differentially reduced in children at high genetic risk for development of atopic disease (30, 52, 53). Additionally, the capacity of neonatal T-cells to efficiently generate immunological "memory" after initial priming is questioned by recent findings (54–56).

Airway intraepithelial dendritic cells have not been examined in human infants, but indirect evidence suggests that the maturation of their counterparts in the gut wall mainly occurs postnatally (57). In rodents, airway intraepithelial dendritic cell populations mature relatively slowly between birth and weaning (58).

Thus, it appears that all of the cell populations involved in the T-cell selection process leading to the generation of immunological memory for inhaled allergens, are undergoing major maturational changes during the key initial stages of this process. This may underlie the increased susceptibility of neonates in general to primary allergic sensitisation, and by extension, delayed maturation of immunocompetence during infancy appears likely to further increase this risk.

Role of environmental factors in the induction of allergic respiratory disease: time for reappraisal?

During the past 20 years, interest in the role of environmental factors has focussed on three major areas, domestic and industrial air pollutants, environmental tobacco smoke (ETS), and respiratory (viral) infections.

The principal literature arguing for a significant role for *air pollutants* comprises population studies demonstrating gradations in the prevalence of allergic disease between rural versus urban and/or industrialised versus non-industrialised populations, and this is supported by data demonstrating increased primary IgE responses in animals exposed acutely to relatively high levels of pollutants, concomitantly with allergen aerosols (e.g. reviews 27, 28). Interpretation of many of the human studies is complicated, however, by the fact that the air pollutants also have the potential to trigger the symptoms of respiratory allergy independent of effects on IgE responses, via direct damage to airway tissue.

Similar considerations cloud the interpretation of data on the effects of tobacco smoke exposure. While active smoking is known to promote primary IgE responses in adults to occupational allergens, where exposure levels of both the allergens and the tobacco smoke are relatively high, there is no convincing evidence that smoking affects responses to "natural" environmental inhalant allergens such as pollen which occur at much lower concentrations in air (59). ETS exposure has likewise been shown to promote occupational allergy in adults (60), and to increase overall serum levels of IgE in children (61); however, its principal role in the development of respiratory allergy symptoms may be by promotion of airways hyperresponsiveness via direct irritant activity (e.g. 62, 63), rather than through effects on the T-cell sensitisation process. It is also of interest to note that it has been clearly established in the immunotoxicology literature that exposure to both industrial air pollutants and tobacco smoke exert effects upon immune responsiveness in inhaled antigens that are biphasic in relation to exposure time and dose, i.e. while short-moderate term exposure to low doses indeed leads to hyper-reactivity, these effects are lost at higher/longer exposures, as the toxic effects of the pollutants become dominant (59, 64, 65).

The role of respiratory infections also requires re-evaluation in this context. It is currently accepted that respiratory viral infections are important "trigger factors" in the development of allergic respiratory diseases, in particular asthma (66). However, as is the case for both ETS and industrial air pollutants, virus infections provide powerful stimuli for the induction of bronchial hyperreactivity via direct

damage to airway tissue, independent of any effects upon IgE responses. The publication universally quoted as evidence for a direct causal link between virus infection and induction of IgE responses to inhalants was based on observations upon 11 children who developed atopic sensitisation diagnosed by RAST, which was preceded by an upper respiratory tract infection (67); this small study dates to 1979, and has not been formally confirmed (footnote at the end of this review).

Recent data emerging from surveys on pediatric populations from eastern and western Europe argue that the role of respiratory infections in this context, are much more complex. Notably, comparative studies in the two Germanys indicate markedly lower prevalence of atopic asthma and sensitisation to inhalant allergens in the East, which is associated with an extremely high prevalence of bronchitis (68, 69); approximately 70% of the East German children were in daily child care by their first birthday compared to 7% in the West, greatly increasing their chances of early respiratory viral infections (69). More detailed studies comparing children in rural and urban Sweden with their counterparts in Poland and Estonia have produced virtually identical conclusions (28, 70) and further provided direct evidence that the decreased prevalence of allergic sensitisation in the former socialist countries is associated with increased prevalence of respiratory infections (70).

Further indication of a potentially "protective" role for infection is provided in recent studies which demonstrate an inverse relationship between allergic sensitisation to inhalants in individual children and number of siblings (71, 72) i.e. children from large families, in which "sharing" of virus infections typically leads to overall higher than average infection rates, display lower than average skin test reactivity. Additionally, the expression of allergic reactivity has also been reported as inversely related to overall socioeconomic status, both in developed countries (73, 74) and in the Third World (2, 75).

It is of interest to note that the air pollution levels in the Eastern European countries studied above are also extremely high, reaching levels accepted in the West as increasing the risk of allergic sensitisation (70). The finding that sensitisation rates are nevertheless extremely low argues either that there are major qualitative differences in pollutants which explain the lack of effects in the East (unconvincing in my view, given that the prevalence in the East is lower than in unpolluted areas in the West), or that the protective effects triggered by infection are sufficiently powerful to negate the weaker (pro-sensitising) signals provided by air pollutants.

A possibility not considered in this discussion is that environmental (especially domestic) levels of

inhalant allergens may be markedly lower in the East than in the West, perhaps as a result of less effective house "sealing" (and hence better ventilation) in the East. No direct data is yet available on this question, but sensitisation patterns argue indirectly against it being a dominant factor (72), particularly for "outside" allergens such as pollens.

Infections and primary allergic sensitisation during infancy: potential mechanisms for infection-mediated "protection"

On the basis of our current perception of how primary IgE responses to inhaled antigens are regulated, it appears feasible that interactions with microbial stimuli could modulate the allergic sensitisation process, on at least two levels.

Microbial stimulation, particularly from the gut flora, provides the principal impetus for normal postnatal maturation of immunoinflammatory function – The newborn of all mammalian species are to varying degrees immunologically "incompetent" at birth, and the underlying cellular function(s) mature postnatally at highly variable rates. As noted above, the capacity to develop protective "tolerance" to both ingested and inhaled allergens is low in rats and mice at birth, and improves progressively during the preweaning period. The epidemiological data linking month of birth with high frequency of sensitisation to seasonal allergens, suggest this transient developmental defect is also shared by humans. This notion is supported by emerging data on the functional status of T-cells and related accessory cell populations in human neonates, summarised above.

What primarily determines the rate of postnatal maturation of these cellular immune function(s)? The answer from the experimental literature is clearly the nature and intensity of microbial stimulation from the environment, in particular the gut flora and its counterpart in the upper respiratory tract. Aside from providing maturation signals for local T-cell-dependent immune function(s) at mucosal surfaces (76), the microbial flora stimulate the functional maturation of the reticuloendothelial system throughout the body, including distal macrophage populations such as those in the peritoneal cavity (77–79) and also in the lung (80–81). Denial (or reduction) of these maturational signals via maintenance of animals in artificial germ free or pathogen-free states markedly inhibits development of both the effector function(s) of macrophage populations (such as mediator secretion), as well as their capacity to respond to activation stimuli (77–81), and would accordingly be likely to reduce their effectiveness in cell-cell interactions such as those described in Fig. 1.

Recent experiments also indicate that the airway intraepithelial dendritic cell population responsible

for acquisition and presentation of inhaled allergen to the T-cell system is exquisitely sensitive to local microbial stimuli in adults (82), and the postnatal maturation of this important cellular network also appears to be "driven" by these same agents (58).

"Bystander" respiratory infections can directly modulate TH1/TH2 selection in parallel immune response to inhaled allergens – As noted above, the scheme in Fig. 1 describes events occurring in the paracortical (T-cell) regions of lymph nodes draining the conducting airways. The initial rate-limiting cells are the dendritic cell population, which is specialised for migration from the airway wall to the node, bearing processed allergen for presentation to resident T-cells. The paracortical regions generally contain few macrophages, and those resident in "upstream" airway mucosal tissues are normally sessile and become migratory only after local inflammatory stimulation; indirect evidence suggests that following mobilisation, those from the lung home selectively to the same paracortical regions in the lymph nodes in which the dendritic cells and T-cells interact (83), placing them at an appropriate location to influence locally developing CD4⁺ T-cell responses as per Fig. 1. Their overall impact on TH-1/TH-2 selection in immune responses to environmental allergens which (as suggested by the data in Fig. 2) evolve via continuous allergenic stimulation over periods of several years, must be directly related to the frequency with which "waves" of these cells are activated and mobilised from the airway wall, and this would in turn be a reflection of infection history.

Conclusions

It may be logically inferred from historical trends in infant mortality and morbidity, that improvements in public health and general living standards in the developed countries over the last 2–3 generations, has progressively reduced the level of exposure of infants and young children to the natural microbial environment, in particular to gastrointestinal and respiratory pathogens. While these measures have had

The precise role of respiratory infection in healthy children in the developed countries cannot, in my view, be accurately assessed in the absence of further detailed epidemiology. It is important to note that the nature of the host response to occasional and (by comparison) relatively short-lived respiratory infections in healthy children living on non polluted environments, may differ both qualitatively and quantitatively from that in East German or Polish children in whom chronic airways inflammation is virtually endemic. It is possible, for example, that the degree of stimulation provided by less frequent/less intense infections in Western children may be insufficient to effectively mobilise the IFN γ /IL-12 pathway described above, thus unmasking other effects of infection (such as increased permeability etc) which may then be read out by the immune system as "TH-2-stimulatory".

clear social benefits, they may additionally have had the less desirable side effect of lowering the average level of microbial stimulation being provided to infants, to a point approaching the critical "threshold" (set via evolution) which is required to optimally drive the final postnatal maturation of key aspects of immunoinflammatory function that are necessary for survival outside the womb. As a result, the kinetics of postnatal development of immune competence has become sluggish, prolonging the period during which the immature immune system is at high risk of making quantitative/qualitative "mistakes", which may subsequently become permanently imprinted in immunological memory. Paramount amongst those immunological "mistakes" appears to be the generation of TH-2 memory against airborne protein allergens.

It is also of interest to note that similar logic is currently being applied in relation to the aetiology of Type I diabetes mellitus, an autoimmune disease which typically manifests in childhood, and which (like atopy) is also increasing steadily in prevalence in developed countries. In this case, the immune system mounts aberrant T-cell mediated cellular immune responses against self antigens expressed on pancreatic islet cells. Clinical trials are in progress in several centres, following on successful pilot studies (84), which are employing non-specific microbial stimulation in children manifesting early stages of the disease, to overcome what is perceived as primarily a deficiency in immunocompetence. It is additionally noteworthy in this context that barrier-housed "specified pathogen free" animals are much more sensitive to the induction of autoimmune insulin-dependent diabetes, than their microbiologically normal "conventionally reared" counterparts (85).

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